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SUICIDE INHIBITORS OF REVERSE TRANSCRIPTASE IN THE

THERAPY OF AIDS AND OTHER RETROVIRUSES

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#### **SUMMARY**

- 1. / The final year of the contract has been quite productive and significant advances have been made in a number of areas.
- 2. Groups of compounds were synthesized as potential suicide inhibitors of viral polymerases. Test results from compounds submitted to the U.S. Army antiviral screening program have been received and are summarized below. Full details of active compounds are given in the Appendix to this report. Of the 40 compounds tested to date, significant antiviral activity against one or more of the 11 test viruses have been observed for a surprisingly high proportion. 16 compounds in all have shown some antiviral activity. Several of these have low cytotoxicity and small animal test are planned in collaboration with the Army antiviral screening program if the current (residual) funding proves sufficient.
- 3. Cellular components have been identified that produce up to a 1,000-fold increase in the sensitivity of HIV-reverse transcriptase to the pyrophosphate analog phosphonoformate (Foscarnet).
- 4. The following table summarizes the compounds found to have antiviral activity and the target virus against which they were effective.

<u>Virus</u>	Com	ounds sh		ificant ant S #'s)	iviral acti	vity
HIV Vaccinia		6460 6462	6457 6467	6455	64 <b>66</b>	
Punta Toro Yellow Fever	6442	6443 6456	6444 6458	6445 6444	6449 644 <b>5</b>	6462

Further details for each compound are given in the Appendix Section.

#### A. INTRODUCTION

The principle that inhibitors of reverse transcriptase will inhibit replication of retroviruses is well established. For example, 3'-azido-3' deoxythymidine, the triphosphate of which inhibits the RT activity of HIV, is a potent inhibitor of virus replication in cultured H-9 cells in the range of 1-10 micromolar and is being used successfully in patients with AIDS. Prolonged administration may cause serious side effects. Another agent which has shown promise is phosphonoformate (PFA, Foscarnet) which inhibits the RT activity of HIV with an I<sub>so</sub> of only 0.1 micromolar. However, much higher concentrations of up to 340 micromolar are required for complete inhibition of HIV replication in H-9 cells. Other drugs including the dideoxycytidines which are also based upon inhibition of RT by chain termination of the viral template are in clinical trial. Since none of these drugs permanently inactivate the reverse transcriptase and since they do not accumulate intracellularly in significant amounts, virus replication will resume when blood levels of the drugs decrease.

This project represents a collaborative effort between groups of investigators with expertise in virology, cell biology, enzymology, drug design and organic synthesis, to develop new types of antiviral drugs. Novel anti-HIV drugs are being developed based upon the principles of (i) compounds designed to accumulate intracellularly at the sites of viral replication (ii) slow release lipid soluble prodrugs having a long biological half life and capability to accumulate in brain tissues (iii) synergistic combination drugs designed to reduce side effects and development of drug resistance during long-term therapy.

#### B. WORK ACCOMPLISHED

#### Cellular Pharmacokinetics of Sterol Phosphonoformates.

The first class of compounds are lipid soluble sterol derivatives of the pyrophosphate analog PHOSPHCNOFORMIC ACID (PFA). This compound is an excellent in vivo inhibitor of reverse transcriptase with I<sub>20</sub>'s as low as 0.1  $\mu$ M for the HIV-RT when transcribing from the viral template. The drug is also relatively non-toxic and acute dosage blood levels of up to 300  $\mu$ M have been reported without serious side effects. The antiviral potency of the compound however is low with concentrations up to 330  $\mu$ M being required for complete inhibition of HIV replication in tissue culture. Simple esterification derivatives do not improve the antiviral potency.

The sterol phosphonoformates which we have developed thus represent a significant advance in the pharmacology of antiviral drug delivery to cells. They are replication-site directed inhibitors designed to enter and accumulate in cells via the endocytotic pathway normally used for cholesterol esters. Subsequent hydrolysis by lysosomal sterol esterases results in slow release of PFA at the intracellular sites where the first critical steps in HIV replication take place. Some of the cholesterol phosphonoformate derivatives we have synthesized display a 20-30 fold increase in potency against

virus replication in tissue culture, compared to the parent compound PFA.

One of the major goals of this project is the further development of this class of compounds into effective therapeutic agents. These studies will include synthesis and evaluation of ligands which enhance cellular accumulation, those which regulate hydrolysis, and those which enhance anti-viral activity against HIV replication in a standardized tissue culture assay system. A novel and potentially useful therapeutic property which has been observed is the ability of these sterol analogs to accumulate intracellularly and protect cells against virus for up to 8 days following drug removal. The pharmacokinetic studies we are proposing on blood-brain and tissue distribution and half life of these compounds in vivo are designed to investigate their suitability as potential agents for longterm antiviral therapy.

#### Sterol Carboxylate Diesters of AZT, DDC and nucleoside spiroxiranes (NSO):

An extension of this strategy which has proved successful with PFA is being applied to improve the pharmacokinetic properties of AZT, dideoxycytidine (DDC) and nucleoside spiroxiranes. The nucleoside spiroxiranes are a new class of mechanism based (suicide) inhibitors of the reverse transcriptase. These nucleoside analogs are effective inhibitors of reverse transcriptase and viral replication but have very short blood half lives and do not accumulate intracellularly. Virus replication probably recovers soon after blood levels of the drugs fall. Here the problem is not to

increase the cellular permeability of the compounds which is adequate, but to enhance the intraceilular accumulation in a slow-release form.

The effect on their pharmacokinetic properties of modifying these compounds by conversion to the 5' sterol dicarboxylates will therefore be investigated. These compounds are designed to incorporate into the lipoprotein and chylomicron particles in the same manner as the long-chain cholesterol esters and phospholipids. Cholesteryl sebacylchloride has been used previously to make synthetic lipoproteins. Sterol and ester hydrolases are present in the lysosomes which will regenerate the active compounds. In pilot studies <sup>3</sup>H-AZT cholesteryl sebacylate has been synthesized and uptake by cultured lymphocytes confirmed (see preliminary data section). Appropriate changes in the sterol and linker moieties will be made and the effects on cell accumulation, intracellular release and prolongation of antiviral protection will be evaluated. In addition, the unlabelled AZT-cholesteryl sebacate, succinate and carbonate esters have been synthesized and inhibited viral replication in tissue culture.

#### 3. Development of Synergistic Combination Drugs:

The basic hypothesis underlying this approach is that drug dosages and side effects can be reduced and antiviral potency increased by suitable combinations of drugs directed at different facets of the viral replicative process. In addition, combination drugs lessen the opportunity for drug resistant variants of the virus to appear because of the low probability of simultaneous mutations against two mechanistically different inhibitors. This approach therefore has required basic information on the effects of different drugs on the various steps involved in intracellular replication of the HIV virus, using the purified HIV-RT to obtain detailed information on the kinetic interactions of the HIV reverse transcriptase with potential inhibitors. As an example of the application of this type of information, the nucleotide and template specificity of the RT has been studied with respect to inhibition by PFA. PFA inhibits only the step in viral replication in which TTP is being incorporated into the viral template. Incorporation of dCTP is relatively insensitive to PFA. Furthermore PFA inhibition is not competitive with respect to TTP for the HIV-RT, indicating that mutations that confer AZT resistance are unlikely to result in co-resistance to PFA.

Part of the antiviral potency of AZT in addition to inhibiting RT has been attributed to its ability to inhibit thymidine kinase, thus lowering intracellular TTP levels. These results taken together suggest that additive or synergistic effects should be observed in joint therapy of the sterol phosphonoformates with AZT, DDC or NSO. Since the side effects of these drugs are directed in part at different facets of cell metabolism whereas the antiviral effects are focussed on the viral reverse transcriptase, a combination of drugs at levels insufficient to impair cell metabolism may nevertheless give complete inhibition of virus replication. The successful development of the sterol dicarboxylates of AZT renders this approach particularly attractive, since all three types of inhibitor

are now potentially available in slow-release and long acting forms.

#### 4. Expression of HiV-Reverse Transcriptase In Different Cell Lines.

In order to determine if the recombinant HIV-reverse transcriptase was expressed in different forms depending upon the cell type, the vaccinia VCF-2! construct was grown in a number of different cell lines of both human, monkey and rodent origin. The expressed reverse transcriptase was tested for inhibition by Foscarnet at two different levels (1 and 10 nanomolar) and compared to the E. Coli recombinant HIV-RT (Kindly donated by Dr. Steven Hughes Fort Detrick M.D.) and the wild type HIV-RT. Both the wild type and E. Coli HIV-RT's were resistant to PFA showing essentially no inhibition at the 10nM level. Previous studies have shown that both enzymes have  $I_{90}$ 's for PFA in the 200-400 nM range. The recombinant HIV-RT expressed in eukaryotic cells however showed a range of phenotypes as indicated in figures 3 and 4 below. Both U-937 and Vero cells expressed enzyme sensitive to 1 nanomolar PFA, whereas human embryo lung and A-498 cells expressed RT-enzyme having wild-type sensitivity. Hela, HuTK- and CV-1 cells as observed previously expressed enzyme having intermediate PFA sensitivity (Figure 1).

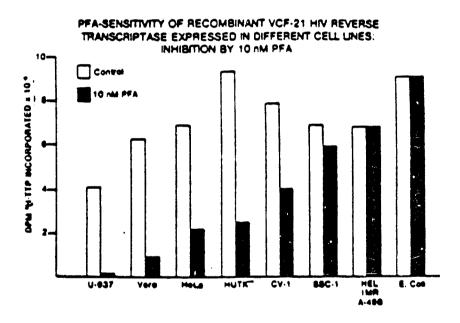
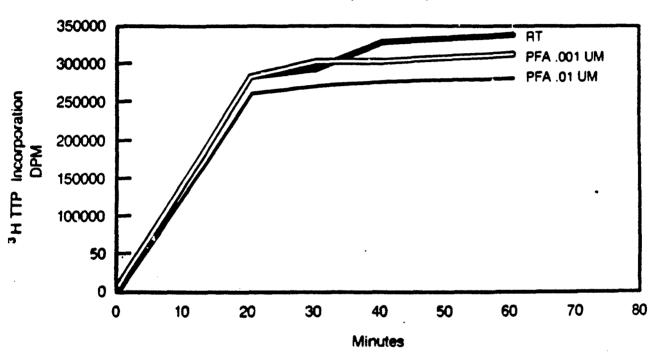


Figure 1: Sensitivity of Recombinant HIV-Reverse Transcriptases to 10 Nanomolar PFA.

The VCF-21 vaccinia construct was grown in the indicated cell lines and the activity of the expressed reverse transcriptase was measured in the presence (dark blocks) or absence (open blocks) of 10 nanomolar PFA.

### 5. Sentivity of Recombinant HIV-Reverse Transcriptase to Foscarnet.

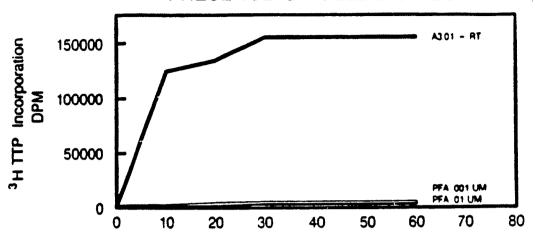
## RECOMBINANT HIV RT (E. COLI) + FOSCARNET



PFA .01 UM

Figure 2. Activity of a recombinant (E. Coli produced) HIV-RT was assayed using  $^3H$ -dTTP and poly rAdT $_{10}$ template both without inhibitor PFA and in the presence of 0.001  $\mu$ M and 0.01  $\mu$ M PFA. Note the relative insensitivity of the enzyme to these low concentrations. The I $_{50}$  of the E. Coli recombinant HIV-RT for PFA was shown to be 0.4  $\mu$ M which is similar to that of the wild type HIV-RT.

# SENSITIVITY TO FOSCARNET INHIBITION OF HIV REVERSE TRANSCRIPTASE (PURIFIED) IN THE PRESENCE OF CELL LYSATES



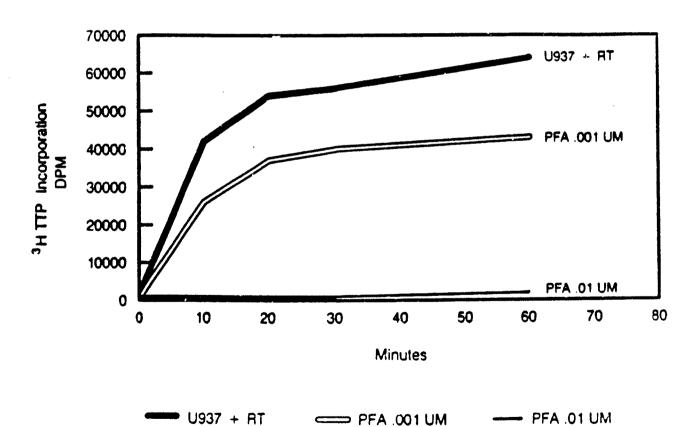


Figure 3. The E. Coli recombinant HIV-RT was incubated with PFA under the same conditions as Figure 1R, with the addition of lysates from A.301 cells (upper panel) or U937 cells (bottom panel). Note that the lysates markedly increase the sensitivity of the reverse transcriptase to inhibition by PFA.

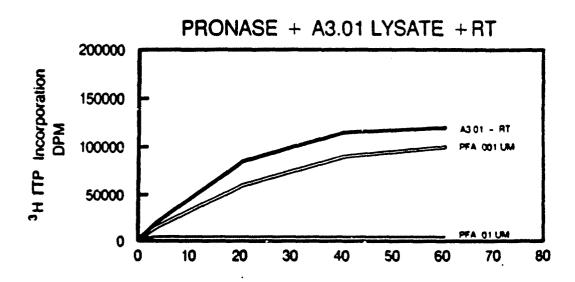
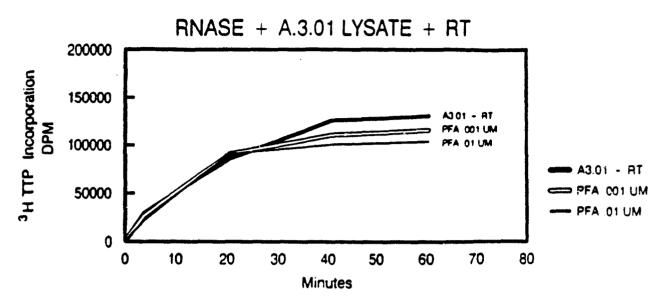


Figure 4. The E. Coli recombinant HIV-RT was incubated with PFA and cell lysates under the same conditions as in Figure 2R with the exception that the lysates were preincubated with pronase for 30 minutes and heat inactivated. Note that neither treatment destroyed the PFA sensitizing activity of the cell extracts.

## SENSITIVITY TO FOSCARNET INHIBITION OF HIV REVERSE TRANSCRIPTASE (PURIFIED) IN THE PRESENCE OF CELL LYSATES



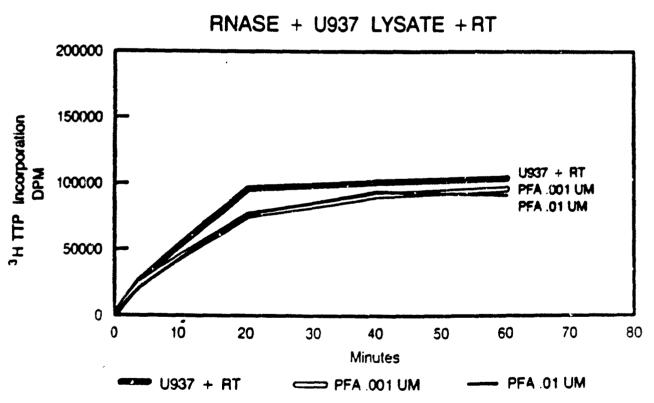


Figure 5. The E. Coli recombinant HIV-RT was incubated with PFA and cell lysates under the same conditions as Figure 2R with the exception that the lysates were preincubated with pancreatic ribonuclease for 30 minutes. Note that the RNAse treatment completely destroyed the PFA sensitizing activity of the extracts.

Since the molecular weight of the sensitized, purified recombinant RT prepared from sensitizing cell lines was not substantially different from that of wild type enzyme, these observations suggest that the sensitizing activity may be associated with a copurifying RNAse sensitive material of relatively low molecular weight.

### 6. Drug Screening Results.

Test results from 26 of the compounds submitted to the U.S. Army antiviral testing facility have been received. A number of compounds were identified that have activity against one or more of the 11 viruses in the test battery. Of these 4 were active against HIV (Appendix III), and 3 were active against Vaccinia. Punta Toro and Yellow Fever viruses (Appendix IV).

active against Vaccinia, Punta Toro and Yellow Fever viruses (Appendix IV).

Several of these compounds have very favorable therapeutic indices and have been selected for further testing and development to the limit of currently available funding.

#### Appendix I.

#### **PUBLICATIONS**

PROJECT: DAMD 17-87-C-7171

TITLE: SUICIDE INHIBITOR OF REVERSE TRANSCRIPTASE IN THERAPY OF AIDS AND OTHER RETROVIRUSES.

PRINCIPAL INVESTIGATOR: Dr. J.M. Bailey, Ph.D., D.Sc. Professor of Biochemistry

#### PRODUCTIVITY REPORT

#### Publications:

- 1. Enhanced sensitivity to Foscarnet of first-strand viral replication by recombinant HIV-reverse transcriptase. M.M. Lightfoote and J.M. Bailey. FASEB J. 4:1318 (1990).
- Differential sensitivity of wild-type and recombinant HIV-Reverse transcriptase to inhibition by Foscarnet. M.M. Lightfoote and J.M. Bailey. Proc V<sup>th</sup> Int. AIDS conf. Montreal 5:515 (1989).
- 3. M.M. Lightfoote and J.M. Bailey. Somatic Cell Modulation of HIV-Reverse Transcriptase Expression. Antiviral Chem. and Chemotherapy. (1989) in preparation.
- 4. Nucleotide and template selectivity for inhibition of reverse transcriptase by PFA: Implications for retroviral therapy. J.M. Bailey and M.M. Lightfoote. *Proc. IV*<sup>th</sup> Int. AIDS Conf. Montreal. 4:3223 (1988).
- 5. Differential sensitivity of wiid-type and recombinant HIV-reverse transcriptase to inhibition by foscarnet. M.M. Lightfoote and J.M. Bailey. *Proc. IV* Int. AIDS Conf. Montreal. (1989).
- 6. Antiviral activities of some sterol phosphonoformate diester. J.M. Bailey, K. Nelson, M. Lightfoote. J. Clin. Exp. Ther. in preparation.
- 7. Nucleoside spiroxiranes: A new class of retroviral inhibitor. J.M. Bailey, K. Nelson, M. Lightfoote. J. Virol. in preparation.
- 8. Synthesis and antiviral activities of some sterol dicarboxylate esters of 3'Azido thymidine (AZT). J.M. Bailey, R.M. Mook, M. Lightfoote. J. Clin. Exp. Ther. in preparation.
- 9. Synthesis of mono and di-substituted cholesterol phosphonoformates by the Arbuzov reaction. J.M. Bailey and Keith Nelson. Tetrahedron Letters. in preparation.

#### Appendix II

#### COMPOUNDS SYNTHESIZED:

#### Compounds synthesized and prepared for shipment to USAMRIID for antiviral testing.

- 1. 2°,02-Anhydrouridine
- 2. 2',0<sup>2</sup>-Anhydrocytidine hydrochloride
- 3. 3',5'-Di-0-benzoyl-2'-02-anhydrouridine
- 4. 5'-0-1-Butyldimethylsilyl-3'-0-benzoyl-2',0'-anhydrouridine
- 5. 2',3'-Anhydro-5'-0-trityluridine
- 6. 3'-Deoxy-2'-thymidinene
- 7. N<sup>3</sup>-Benzyl-2',5'-di-0-trityluridine
- 8. 5'-0-t-Butyldimethylsilylanhydrouridine
- 9. N<sup>4</sup>-Benzoyleytidine
- 10. 2',3'-Di-0 -mesyl-5'-0-trityluridine
- 11. 5'-0 -t-Butyldimethylsilyl-2',3'-isopropylideneuridine
- 12. 2',3'-Isopropylideneuridine
- 13. 2',3'-0-Sulfinylurid' ?
- 14. 2',3'-Benzylideneuridine
- 15. N<sup>4</sup>-Benzoyl-2',3'-0-Sulfinylcytidine
- 16. 2',3'-0-Sulfinylcytidine
- 17. 3',5'-Di-0-trityl-2'deoxy-2'-oxouridine
- 18. 3'.5'-Di-0-t-butyldimethylsilyl-2'-deoxy-2'-oxouridine
- 19. 2'.5'-Di-0-t-butyldimethylsilyl-3'-deoxy-3'-oxouridine
- 20. Diethyl (cholesteryloxycarbonyl) phosphonate
- 21. Disodium (cholesteryloxycarbonyl) phosphonate
- 22. Di-[1-(3-carboethoxypropyl)] cholesteryloxycarbonyl
- 23. Di-(2,3-isopropylideneglyceryl) cholesteryloxycarbonyl phosphonate
- 24. Di-[1-(3-methylbutyl)] cholesteryloxycarbonyl phosphonate Di-[1-(lithium 3-carboxypropyl)] cholesteryloxycarbonyl phosphonate
- 25. Sodium ethyl (cholesteryloxycarbonyl) phosphonate

- 26. Sodium 1-(3-carboxypropyl) 1-(30 carboethoxypropyl) [cholesteryloxycarbonyl] phosphonate
- 27. Adenosine 2',3'-Riboepoxide
- 28. Thymidine 5'-(1,3,2-dioxaphosphorin-2-oxide)
- 29. Thymidinene 5'-(1,3,2-dioxaphosphorin-2-oxide)
- 30. Thymidinene
- 31. 2-Ethoxy-5-chloro-6-methyl-1,3,2-dioxaphosphorin-5-ene-2-oxide
- 32. 2-Ethoxy-5-chloro-1,2-oxaphosphol-4-ene-2-oxide
- 33. 2,4-dichloro-5-methyl-1,3,2-dioxaphosphole-2-oxide
- 34. 2-methoxy-4,5-dimethyl-1,3,2-dioxaphole-2-oxide
- 35. Thymidine 3',5'-oxetane

Appendix III

TEST DATA ON ANTI HIV DRUGS



#### DEPARTMENT OF THE ARMY

ILS. ARMY MEDICAL RESEARCH RISTITUTE OF INFECTIOUS DISEASES
FORT DETRICK, PREDEFICK, MARYLAND 21701-8915

May 10, 1990

REPLY TO

Department of Antiviral Studies

Dr. J. Martyn Bailey
Professor of Biochemistry and Molecular Biology
The George Washington University
Department of Biochemistry
2300 Eye Street, NW
Washington, DC 20037

Dear Dr. Bailey:

Enclosed please find results of the antiviral activity screening on the U.S. Army's Antiviral Drug Development Program. The enclosed data summarizes the *in vitro* results of the screening done to date.

\*Older assay methodologies have been reviewed in relation to current program status and predictability. As a result of this review, current data sheets may reflect new data as well as the removal of previously reported data now thought to be supplanted by new assay techniques.

\*Data have not been previously reported for compounds showing antiviral activity prior to confirmation. I feel this procedure has unnecessarily slowed the reporting of data to suppliers; hence, we will now report data as it is received. Please do not make corporate or business decisions based on a preliminary, unconfirmed result without discussing this data with the undersigned or a designated member of the Virology Division.

Our intent with these changes is to decrease the length of time required to get data to you for review. Please let me know if the new approach to reporting is improved, and if additional modifications might further enhance collaboration.

Correspondence regarding the evaluation of your compounds or the interpretation of screening results should be addressed to the undersigned at U.S. Army Medical Research Institute of Infectious Diseases, Fort Detrick, Frederick, Maryland 21701-5011, (301) 663-7494, FAX (301) 698-0854. Alternatively, you may contact Dr. Edward L. Stephen, P.O. Box 248, Monrovia, Maryland 21770, (301) 874-5533.

Sincerely,

John W. Huggins, Ph.D.

Department of Antiviral Studies

• \_ ′

Virology Division

#### Enclosures

CF: Edward L. Stephen, D.V.M.
Antiviral Information, Compound
Solicitation and Repository

### USAMRIID Antiviral Drug Screening Program SUBMITTER CTR NO AVS NO STRUCTURE CHIRAL 01141.01 KN-11-55 AVS-006466 DATE RECD AMT RECEIVED [mg] MOL WT (au) 12-28-89 53.30 224.213 HANDLING/STORAGE NH SOLUBILITY STABILITY ALT NAME 2',3'-DIDEOXYTHYMIDINENE COMPOUND NAME 2', 3'-DIDEOXYTHYMIDINENE SCREEN INSTRUCTION IN VIVO TOXICITY [mg/kg] PRIORITY=PT>VEE>YF>KHF>PIC>JE>SF>VV>AD2>VSV IN VITRO SCREEN [ug/ml] IN VIVO SCREEN [Dose = mg/kg] SO MTT04-APR-90 SO MTT24-APR-90 SO MTT06-MAR-90 SO MTT06-MAR-90 SO MTT06-MAR-90 SO MTT06-MAR-90 SO MTT06-MAR-90 HTC 66.4 71.1 184 182 171 VIR HST VR VR+ DOSE MTG VEH RTE D TOX SP L PR DATE 4.99 .32 NOT ACT NOT ACT NOT ACT NOT ACT HT2 CEM VERO VERO VERO VERO VERO 13.29 HIV HIV JE PT SP VEE YF > 320 173

PLATE 1Q9 DRUG 6460

# IN VITRO ANTIVIRAL RESULTS MTT ASSAY

DRUG: AVS 6460 TAI: >30.24 SI: 2.79

1	2	3	4	5	6	7	8	9	10	11	12
		reagent basi	tereund					-	reund		
0.302	0.273	0.281	0.414	0.402	0.296	0.0/9	0.081	0.255	0.252	0.130	0.0/1
	***					tox	drug	6460 capery	nontel	00/10	104
: 1	1.940			I		1.999	0.798	0.734	0.680	2.096	2.160
1	1.881			1		1.993	0.697	0.802	0.720	1.992	1.693
1 (	1.776			İ		1.987	0.947	0.994	0.879	1.856	1.943
	0.557			- 1		2.067	1.159	0.949	0.911	0.423	2.060
1 1	0.423					2.018	0.930	1.338	1.309	0.365	1.921
	0.478			i		1.223	0.719	0.614	0.637	0.500	0.881
									erimetria beo	Egraund	
						0.354	0.270	0.282	0.268	0.273	0.313
1300-00 <b>0</b> to	resulty co-	iorinos ber	VO-VIVUS OB!	1 trad	800	- highest dru	g cone		values sho	un are option	denetties

VIPIUS CELLS Shipment number Strn	HIVCRIF CEM 63 RF2	Satisfactory: Active: Retest	PROJECT # SPONSOR TEST DATE DATE READ	6520-2 USAMRIID 06/12/90 06/19/90	
REAGENT	0.328	ERDR:6488:		954	
VIRUS CONTROL CELL CONTROL DIFFERENTIAL	0.130 1.596 1.466	TC (UG/M:): E2:A5:	(数:20) 22.40 4.14	> 100.00	

D	RUG	6460	ANTIVIRAL	TEST VALUES	CYTOTOXICI		
ROW PL	ATE	CONC. (uG/mL)	HEAN O.D.	* RED. IN CPE	MEAN 0.D.	VIABILITY	COLORIMETRIC CONTROL
low	B	0.32	0.295 0.337	204 234	1.767 1.570	1004 984	015
	ŏ	3.2	0.542	374	1.697	1004	055 060
	E	10	0.595	419	1.782	1004	046
high	6 4	32 100	0.793 0.173	549 129	1.700 0.698	1004 444	058 0.026

#### values sharm are final adjusted numbers

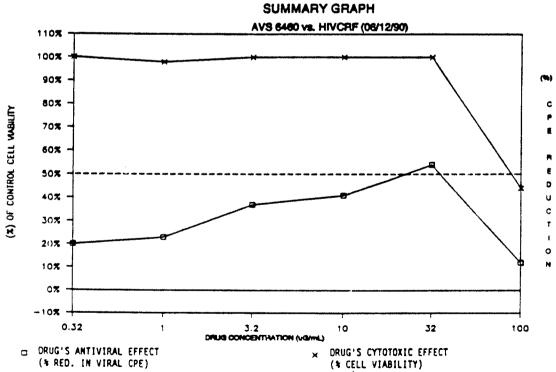


PLATE 1Q8 **DRUG 6457** 

### IN VITRO ANTIVIRAL RESULTS MTT ASSAY

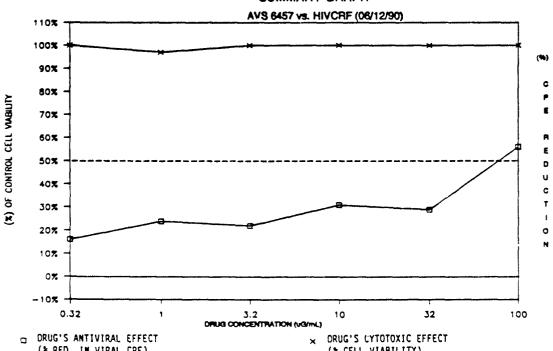
**DRUG: AVS 6457** TAI: >27.78 SI: >1.29

	1	2	3	4	5	6	7	8	9	10	11	12
ſ			reagent best	ground					plants backs	round		
A	0.433	0.373	0.552	0.655	0.447	0.451	0.103	0.100	0.191	0.442	0.104	0.087
Γ	144	***	drug	6467 experts	nental	YOU .					08/46	
8	1.739	1.747	0.707	0.601	0.807	1.972				!	1.841	
c	1.606	1.765	0.674	0.715	1.069	1.794	]			1	1.750	
0	1.871	1.913	0.55\$	0.799	1.037	1.806					2.012	
E	1.782	0.522	0.965	0.990	0.742	1.916				- 1	0.811	
F	1.783	0.541	0.894	0.662	1.126	1.899	1			•	0.583	
6	1.691	0.602	1.069	1.161	1.241	1.811	1			1	0.652	
ſ		drug 8157 exteriments background										
H	0.345	0.405	0.387	0.396	0.393	0.382						
•		4				200						

VIRUS CELLS SHIPHENT NUMBER STRN	HIVCRF CEM 63 RF2	Satisfactory: Active: Retest	PROJECT # SPONSOR TEST DATE DATE READ	6520-2 USAHRIID 06/12/90 06/19/90
REAGENT	0.485	ORISC 6457 254	504×	954
VIRUS CONTROL CELL CONTROL DIFFERENTIAL	0.133 1.353 1.220	TC (UG/ML) > 180.08 180.08 4.68	> 190.00 77.60 > 1.29	> 100.00

	TEST VALUES	CYTOTOXICIT	TEST VALUES	DRUG 6457		
COLOD VETRIC	* CELL VIABILITY	HEAN O.D.	4 RED. IN VIRAL CPE	MEAN O.D.	CONC. (uG/mL)	ROW ON PLATE
092 089 098	1004 974 1004 1004 1004 1004	1.473 1.307 1.442 1.462 1.436 1.406	164 244 224 314 294 564	0.190 0.293 0.269 0.379 0.356 0.679	0.32 1 3.2 10 32 100	low 8 C D E F high G *

### **SUMMARY GRAPH**



(% RED. IN VIRAL CPE)

(\* CELL VIABILITY)

PLATE 1Q7 DRUG 6455

### IN VITRO ANTIVIRAL RESULTS MTT ASSAY

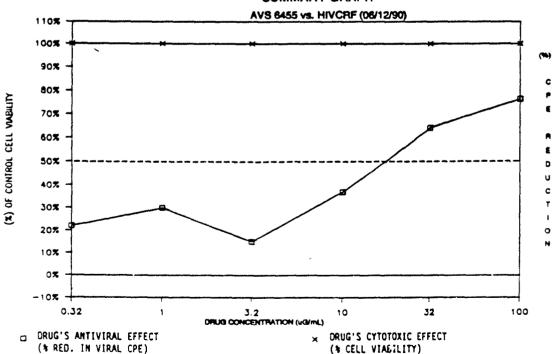
**DRUG: AVS 6455** TAI: >38.98 SI: >5.71

_	1	2	3	4	5	6	7	8	9	10	11	12
. i			respont box	-					homen news	-		
A L	0.463	0.349	0.539	0.566	0.386	0.388	0.253	0.256	0.399	0.448	0.239	0.199
_ [	Will I	88/10		GAGE experie		VOSE .					00/10	
B	1.916	1.590	0.861	0.677	0.813	1.889				1	1.783	
C j	1.340	1.486	0.820	0.986	0.873	2.017	· • ]			J	1.588	
0	1.800	2.193	0.764	0.557	0.841	1.849				Į.	1.641	
E	2.057	0.462	0.856	0.811	1.334	2.016	[				0.539	
F	1.959	0.636	0.944	1.071	1.242	1.830	1				0.548	
6	2.003	0.374	1.269	1.478	1.180	1.913					0.607	
ſ			drug BASS to	iarlmetria bac	icground							
H	0.328	0.243	C.480	0.459	0.455	0.448						
_	100-006			-		act D	- Nichass day	-		maken abou		december

VIFIUS CELLS SHIPMENT NUMBER STRN	HIVCRIF CEMI 63 RF2	Satisfactory: Active: Retest	PROJECT # SPONSOR TEST DATE DATE READ	6520-2 USAMRIID 06/12/90 06/19/90	
REAGENT	0.449	DRUG 6455 254	504	954	
VIRUS CONTROL CELL CONTROL DIFFERENTIAL	0.079 1.265 1.186	TC (UG/ML) > 100.00 IC (UG/ML) 0.49 ANTIVIRAL INDEX (AI) > 203.84	> 100.06 17,50 > 5.71	> 100.00	

	TY TEST VALUES	CYTOTOXICI	TEST VALUES	ANTIVIRAL	DRUG 6455	
COLORIMETRIC CONTROL	* CELL VIABILITY	MEAN O.D.	* RED. IN VIRAL CPE	MEAN 0.D.	CONC.	ROW ON PLATE
0.000 0.007 0.011 0.031 206 120	1004 1004 1004 1004 1004 1004	1.454 1.473 1.365 1.557 1.652 1.631	224 304 154 374 644 764	0.256 0.358 0.182 0.442 0.764 0.902	0.32 1 3.2 10 32 100	low B C D E F high G **

#### **SUMMARY GRAPH**



(\* CELL VIAGILITY)

SOUTHERN RESEARCH INSTITUTE

# Appendix IV TEST DATA AGAINST OTHER VIRUSES

## USAMRIID

TRUCTURE	iral Drug Scr chiral	SUBMITTER	CTR NO	03/26/9 AVS NO
	CHINAL	01141.01	KN-V-99	AVS-006442
		DATE RECD	AMT RECEIVED [mg]	
° 		12-28-89	74.00	290.253
NH NH		HANCLING/STORAGE		
( j				
0 0	၁			
но		SOLUBILITY		
° , °		STABILITY		
!! •				
•		ALT NAME	<u> </u>	
		ì	,3'-0-SULFINYL URIDI	NE
MPOUND NAME		1		
	2', 3'-0-SULFIN	IYL URIDINE		
SCREEN INSTRUCTIO	ON .	IN V	IVO TOXICITY [n	ig/kg]
ORITY=PT>VEE>YF>KHF>PIC>JE>SF>VV>AD2			SO HTC LAB PR DATE	
		1044, 76 5.5	ALC HOLE TO BOLD	
	, , , , , , , , , , , , , , , , , , , ,			
IN VITRO SCREEN (ug	/ml]			mg/kg]
VR VR+ LD50 CELL MTC TI HOT ACT VERO 183 0	TI: LAB PRT DATE SO HTT 90-03-01		POSE HTC VEH RTE D TOX S	P L CR CATE
100 VERO 170 2.39 MOT ACT VERO 172 0 MOT ACT VERO 38.3 0	00-50-01 TTM OS SO MTT 90-03-01 SO MTT 90-03-02			
NOT ACT VENO 171 0	SO MTT 90-03-01			
	•			
		1		
		1		

PLATE U9A

# IN VITRO ANTIVIRAL RESULTS MTT ASSAY

DRUG: AVS 6442 TAI: 15.38 SI: 1.70

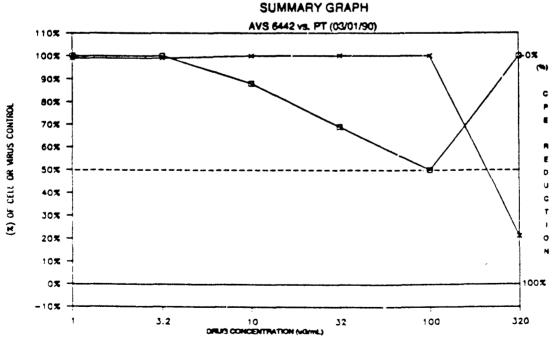
	1	2	3	4	5	6	7	•	•	10	11	12
ſ			-	egraund .		T	·		-	raund		
A	0.042	0.041	0.040	0.042	0.039	0.042	0.001	0.001	0.001	0.001	0.001	0.001
1	•== 1		drug	6442 emerir	non tel	YOM					00/100	
•	0.898	0.951	0.392	0.299	0.337	0.956					0.746	
c	0.930	1.033	0.389	0.295	0.345	0.912	j			1	0.972	
ا ۵	1.011	1.139	0.419	0.423	0.434	1.020	1				0.800	
	1.034	0.329	0.559	0.504	0.542	1.102	1				0.334	
	1.115	0.389	0.663	0.626	0.656	1.181	1				0.342	
•	0.243	0.553	0.227	0.227	0.219	0.229	1				0.396	
- 1			<b>6449 6443 00</b>	-	refround							
. ■	0.049	0.038	0.038	0.037	0.036	0.039						
•	-											1.40000000

VIRUS CELLS SELFIGUE KAGER STIS	PT VERO 63 AGAMES	Satisfactory: Active: Ret	PROJECT # SPONSOR TEST DATE DATE READ	5975-1 USAMRIID 03/01/90 03/09/90	
KEAGEN?	0.041	DRUG 6442	25%	50%	95%
VIRUS CONTROL	0.316	TC (wG/mL)	170.00	239.00	> 320.00
CELL CONTROL	0.899	IC (wd/mL)	22.20	100.00	
DIFFERENTIAL	0.583	ANTIVIRAL INDEX (AI)	7.65	2.39	

DRUG		6442	ANTIVIRAL I	EST VALUES	CALOLOXICI	IY TEST VALUES		
ROW	90	COMC.	HEAM	9 VIRAL	POEAUE	1 CELL	CLURINGTRIC	
PLATE		(uG/mL)	0.0.	CPE	0.0.	VIABILITY	CONTROL	
10-	3	1	013	1004	0.488	994	002	
	c	3.2	011	1009	0.887	999	003	
	D	10	0.072	400	0.979	100	004	
	1	32	0.161	694	1.030	1004	003	
	7	100	0.294	509	1.110	100	003	
igh	4	320	141	100	0.187	219	0.000	

\* Nighest Grug eancentration temed

values shown are final adjusted numbers



G DRUG'S AMTIVITAL EMMECT
(% VIRAL CPE)

X DRUG'S CYTOTOXIC EFFECT
(% CELL VIABILITY)

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SOUTHERN RESEARCH INSTITUTE

#### USAMRIID

Antiviral Drug Screening Program SUBMITTER 01141.01 AVS NO CTR NO TRUCTURE CHIRAL KN-V-109 AVS-006443 DATE RECD AMT RECEIVED [mg] MOL WT (au) 12-28-89 818.979 86.00 HANDLING/STORAGE SOLUBILITY STABILITY ALT NAME N3-BENZYL-2',5'-CI-O-TRITYLURIDINE 120UND NAME N3-BENZYL-2',5'-DI-O-TRITYLURIDINE SCREEN INSTRUCTION IN VIVO TOXICITY [mg/kg] [ORITY=PT>VEE>YF>KHF>PIC>JE>SF>VV>AD2>VSV IN VITRO SCREEN [ug/ml] IN VIVO SCREEN [Dose = mg/kg] VIR HST VR VR+ DOSE HTC VEH RIE D TOX SP L PR DATE 50 MTT 90-03-01 50 MTT 90-03-01 50 MTT 90-03-01 50 MTT 90-03-02 50 MTT 90-03-01 VERO VERO VERO VERO 24.7 210 > 320 > 320 > 320

PLATE U9A DRUG 6443

# IN VITRO ANTIVIRAL RESULTS MTT ASSAY

DRUG: AVS 6443 TAI: >10.57 SI: 2.72

0.042	0.041	•	graund													
0.042	0.041		reagent background						ent background plants background							
		0.040	0.042	0.039	0.042	0.001	0.001	0.001	0.001	0.001	0.001					
1	00/16					104	drug	6443 experve	nental	00/10	tos					
- 1	0.951			1		0.830	0.376	0.357	0.334	0.746	0.879					
1	1.033			1		0.911	0.386	0.324	0.297	0.972	0.77					
L	1.139					1.015	0.406	0.436	0.343	0.800	0.86					
	0.329			- 1		0.800	0.493	0.491	0.497	0.334	0.81					
1	0.389			1		0.734	0.695	0.713	0.683	0.342	0.71					
	0.353			1		0.696	0.560	0.632	0.599	0.396	0.75					
						Ī		1749 BUT	iarmetris bea	eground						
						0.059	0.044	0.040	0.039	0.040	0.040					
_		1.033 1.139 0.329 0.389 0.353	1.033 1.139 0.329 0.369 0.353	1.033 1.139 0.329 0.369 0.353	1.033 1.139 0.329 0.389 0.353	1.033 1.139 0.329 0.389 0.353	1.033 1.139 0.329 0.389 0.353 0.059	1.033 1.139 0.329 0.389 0.353 0.059 0.059 0.059 0.044	1.033 1.139 0.329 0.329 0.389 0.353 0.353 0.696 0.560 0.696 0.059 0.044 0.040	1.033 1.139 1.035 0.329 0.329 0.389 0.353 0.353 0.696 0.560 0.696 0.696 0.696 0.049 0.049 0.059 0.044 0.049 0.039	1.033 1.139 1.015 0.406 0.324 0.297 0.972 1.139 1.015 0.406 0.436 0.343 0.800 0.329 0.389 0.389 0.394 0.695 0.713 0.693 0.342 0.353 0.696 0.560 0.632 0.599 0.396  drug 6443 coordinates background 0.059 0.044 0.040 0.039 0.040					

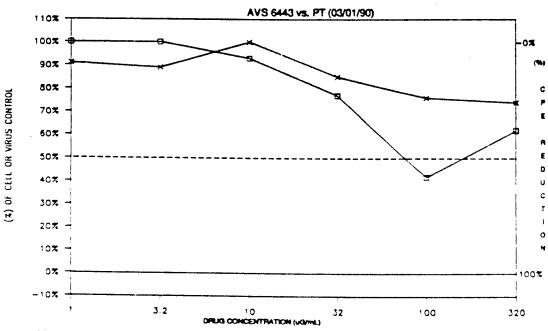
VIRUS	PT			770	JECT (	597	5-1
CELLS	VERO	Satisfactory; Active; Ret	est	570	HSOR	USAMRIID	
SHIPHENT NUMBER	63			TES	T DATE	03/	01/90
STRU	ADAMES			DAT	E READ	03/	09/90
reagent	0.041	DRUG 6443	25℃		50%		95%
VIRUS CONTROL	0.316	TC (uG/mL)	210.00	>	320.00	>	320.00
CELL CONTROL	0.899	IC (ue/mL)	34.20		77.19		
DIFFERENTIAL	0.583	MYTIVIRAL INDEX (AI)	6.15	>	4.15		

DRUG 6443		6443	ANTIVIRAL T	EST VALUES	CYTOTOXICI		
ROW	OM	CONC.	MILAN	♦ VIRAL	KEAN	♦ CELL	COLORIDETRIC
PL	ATT	(uG/mL)	0.0.	CPE	0.0.	VIABILITY	CONTROL
10₩	3	1	001	100%	0.815	91%	001
	C	3.2	021	100	0.801	897	001
	D	10	0.040	934	0.902	100	002
	I	32	0.137	77%	0.767	05%	001
	7	100	0.337	429	0.601	769	0.003
nigh	G :	320	0.222	621	0.665	749	0.010

\* highest drug concentration leases

values shown are final adjusted number

# SUMMARY GRAPH



DRUG'S ANTIVIRAL EFFECT
(% VIRAL CPE)

x DRUG'S CYTOTOXIC EFFECT
{\\$ CELL VIABILITY}

USAM	RIID
Antiviral Drug Scr	reening Program
STRUCTURF CHIRAL	SUBMITTER CTR NO AVS NO 01141.01 KN-VII-83 AVS-006444
	DATE RECD ANT RECEIVED [mg] MOL WT (al) 12-29-89 79.00 726.837
NH NH	HANDLING/STORAGE
	SOLUBILITY
	STABILITY
	ALT NAME  3'-DEOXY-2',5'-DI-O-TRITYL-3'-OXOURIDINE
COMPOUND NAME  3'-DEGXY-2',5'-DI-O-TR	ITYL-3'-OXCURIDINE
SCREEN INSTRUCTION	IN VIVO TOXICITY [mg/kg]
PRIORITY-PT>VEE>YF>KHF>PIC>JE>SF>VV>AD2>VSV	HOST VH RTE LOSS MTC LAB PR DATE
IN VITRO SCREEN [ug/ml]	IN VIVO SCREEN [Dose = mg/kg]
VIR VR VR- (250 CELL HTC 1; TI- LAB PRT DATE	VIR HET VR VR- DOSE HTG VEH RTE D TOX SP L PR DATE
TE	

~ •

PLAT	E	U96
A PARTICULAR A	4	

#### IN VITRO ANTIVIRAL RESULTS MTT ASSAY

#### **DRUG: AVS 6444** TAI: >24.65 SI: >4.04

_	1	2	3	4		•	7		9	10	11	12
Г		1	vagent basi	-		i			places seeing	round		
A	0.043	0.042	0.045	0.041	0.040	0.042	0.001	0.001	0.001	0.001	0.001	0.001
Г	100	00/10	drug	\$444 expense	Yen Lai	10%	i				00/740	
3	1.033	0.918	0.365	0.330	0.321	0.864				1	0.837	
c	0.959	0.946	0.304	0.333	0.381	0.999	1			ļ	0.734	
0	0.936	1.054	0.433	0.451	0.414	0.883	i			[	0.631	
	0.011	0.295	0.382	0.411	3.490	0.810	1				0.339	
7	0.753	0.355	0.577	0.706	0.655	0.748	- [			į	0.369	
a [	0.902	0.302	0.952	0.924	0.879	0.981					0.350	
Γ			drug 6444 ap	istractic bas	egraund							
	0.041	0.043	0.042	0.039	0.039	0.039						
_	Managel to	HOLEN OF	-	-	100	8010	- highest dr	4 0004		Values sho		deneries

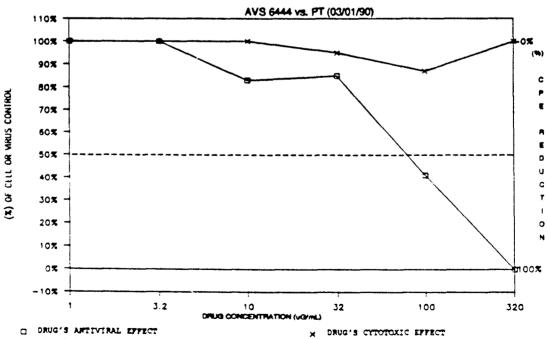
VIRUS CELLS SEITHERT HUGER STRE	PT VERO 63 ADARES	Satisfactory: Active: R	etest	PROJECT # SPONSOR TEST DATE DATE READ	5975-1 USAMRIID 03/01/90 03/09/90
REAGENT	0.042	DRUG 6444 25%		50%	95%
VIRUS CONTROL	0.300	TC (ug/mL)	> 320.00	> 320.00	> 320.00
CELL CONTROL	0.811	IC (ve/mL)	41.50	79.20	278.00
DIFFERENTIAL	0.504	AMPINIBAL INDEX (AI)	> 7.72	> 4.04	> 1.15

DRU	0 6444	ANTIVIRAL T	EST VALUES	CALOLOXICI			
NOW C	M CONC.	HEAH	* VIRAL	KELLIK	# CELL	COLORIDETRIC	
PLAT	2 (ud/sL)	0.0.	CPE	0.0.	VIABILITY	CONTROL	
low I	1	008	100	0.909	100%	003	
c	3.2	007	1000	0.940	1000	003	
0	10	0.086	639	0.870	100%	003	
1	32	0.078	05%	0.768	95%	2.000	
,	100	0.295	419	0.707	879	0.001	
aigh d	320	0.570	0	0.940	100%	001	

\* Nighost drug consentration tested

values shown are final adjusted numbers

#### **SUMMARY GRAPH**



(% VIRAL CPE)

(% CELL VIABILITY)

USAM	RIID
Ambiguinal Dunca Con	usasias Dusamas
Antiviral Drug Sci	SUBMITTER CTR NO AVS NO
china china	01141.01 KN-VII-21 AVS-006445
	DATE RECD AMT RECEIVED [mg] MOL WT (au)
	12-28-89 74.00 726.837
	HANDLING/STORAGE
NH	Invited Line of Colored
	SOLUBILITY
• ""	SOLUBILITY
	STABILITY
_	ALT NAME
	2'-DEOXY-3',5'-DI-O-TRITYL-2'-OXOURIDINE
AME STATE OF THE S	TAME OF CAMPARATE
2'-DEOXY-3',5'-DI-O-TR	HITTL-27-OXOURIDINE
SCREEN INSTRUCTION	IN VIVO TOXICITY [mg/kg]
IORITY=PT>VEE>YF>KHF>PIC>JE>SF>VV>AD2>VSV	HOST VM RTE LDSO MIC LAB PR DATE
	1793
IN VITRO SCREEN [ug/ml]	
	IN VIVO SCREEN [Dose = mg/kg]
	IN VIVO SCREEN [Dose = mg/kg]
NOT ACT   VENO   23.2 0   SO HIT 90-03-0	VIA HST VR VR+ DOSE HTC VCH RTE D TOX SP L PR DATE
VR VR+   D50   CELL   MTC   T    T +   LAB   PRT   CATE	VIR HST VR VR+ DOSE HTC VCH RTE D TOX SP L PR DATE
NOT ACT VERO 23.2 0 SO MTT 90-03-0 NOT ACT VERO 23.2 0 SO MTT 90-03-0 22.6 VERO 49 2.92 SO MTT 90-03-0 NOT ACT VERO 43.3 0 SO MTT 90-03-0	VIA HST VR VR+ DOSE HTC VCH RTE D TOX SP L PR DATE
VR VR+   ID50   CELL   HTC   T    T +   LAB   PRT   CATE	VIA HST VR VR+ DOSE HTC VUH RTE D TOX SP L PR DATE
VR VR* 1050 CELL MTC TI TI* LAB PRT CATE  NOT ACT VERO 23.2 0 50 MTT 90-03-0  22.6 VERO 49 2.92 50 MTT 90-03-0  NOT ACT VERO 43.3 0 50 MTT 90-03-0  NOT ACT VERO 32 0 50 MTT 90-03-0	VIA HST VR VR+ DOSE HTC VUH RTE D TOX SP L PR DATE
VR VR* 1050 CELL HTC TI TI* LAB PRT CATE  NOT ACT VERO 23.2 0 50 HTT 90-03-0  22.6 VERO 49 2.92 50 HTT 90-03-0  HOT ACT VERO 43.3 0 50 HTT 90-03-0  NOT ACT VERO 32 0 50 HTT 90-03-0	VIA HST VR VR+ DOSE HTC VUH RTE D TOX SP L PR DATE
VR VR+ 1050 CELL MTC TI TI+ LAB PRT CATE  NOT ACT VERO 23.2 0 SO MTT 90-03-0 22.6 VERO 49 2.92 SO MTT 90-03-0 NOT ACT VERO 43.3 0 SO MTT 90-03-0 NOT ACT VERO 32 0 SO MTT 90-03-0 NOT ACT VERO 32 0 SO MTT 90-03-0	VIA HST VR VR+ DOSE HTC VUH RTE D TOX SP L PR DATE
VR VR* 1050 CELL MTC TI TI* LAB PRT CATE  NOT ACT VERO 23.2 0 50 MTT 90-03-0  22.6 VERO 49 2.92 50 MTT 90-03-0  NOT ACT VERO 43.3 0 50 MTT 90-03-0  NOT ACT VERO 32 0 50 MTT 90-03-0	VIA HST VR VR+ DOSE HTC VCH RTE D TOX SP L PR DATE
VR VR+ 1050 CELL MTC TI TI+ LAB PRT CATE  NOT ACT VERO 23.2 0 SO MTT 90-03-0 22.6 VERO 49 2.92 SO MTT 90-03-0 NOT ACT VERO 43.3 0 SO MTT 90-03-0 NOT ACT VERO 32 0 SO MTT 90-03-0 NOT ACT VERO 32 0 SO MTT 90-03-0	VIA HST VR VR+ DOSE HTC VCH RTE D TOX SP L PR DATE
VR VR* 1050 CELL HTC TI TI* LAB PRT CATE  NOT ACT VERO 23.2 0 50 HTT 90-03-0  22.6 VERO 49 2.92 50 HTT 90-03-0  HOT ACT VERO 43.3 0 50 HTT 90-03-0  NOT ACT VERO 32 0 50 HTT 90-03-0	VIA HST VR VR+ DOSE HTC VUH RTE D TOX SP L PR DATE
VR VR* 1050 CELL MTC TI TI* LAB PRT CATE  NOT ACT VERO 23.2 0 50 MTT 90-03-0  22.6 VERO 49 2.92 50 MTT 90-03-0  NOT ACT VERO 43.3 0 50 MTT 90-03-0  NOT ACT VERO 32 0 50 MTT 90-03-0	VIA HST VR VR+ DOSE HTC VCH RTE D TOX SP L PR DATE
VR VR+ 1050 CELL HTC TI TI+ LAB PRT CATE  NOT ACT VERO 23.2 0 SO HTT 90-03-0 22.6 VERO 49 2.92 SO HTT 90-03-0 NOT ACT VERO 43.3 0 SO HTT 90-03-0 NOT ACT VERO 32 0 SO HTT 90-03-0 NOT ACT VERO 32 0 SO HTT 90-03-0	VIA HST VR VR+ DOSE HTC VCH RTE D TOX SP L PR DATE
VR VR+ 1050 CELL MTC TI TI+ LAB PRT CATE  NOT ACT VERO 23.2 0 SO MTT 90-03-0 22.6 VERO 49 2.92 SO MTT 90-03-0 NOT ACT VERO 43.3 0 SO MTT 90-03-0 NOT ACT VERO 32 0 SO MTT 90-03-0 NOT ACT VERO 32 0 SO MTT 90-03-0	VIA HST VR VR+ DOSE HTC VUH RTE D TOX SP L PR DATE
VR VR* 1050 CELL MTC TI TI* LAB PRT CATE  NOT ACT VERO 23.2 0 50 MTT 90-03-0  22.6 VERO 49 2.92 50 MTT 90-03-0  NOT ACT VERO 43.3 0 50 MTT 90-03-0  NOT ACT VERO 32 0 50 MTT 90-03-0	VIA HST VR VR+ DOSE HTC VUH RTE D TOX SP L PR DATE
NOT ACT VERO 32 0 SO HTT 90-03-0  NOT ACT VERO 49 2.92 SO HTT 90-03-0  NOT ACT VERO 43.3 0 SO HTT 90-03-0  NOT ACT VERO 32 0 SO HTT 90-03-0  SO HTT 90-03-0  SO HTT 90-03-0	VIA HST VR VR+ DOSE HTC VUH RTE D TOX SP L PR DATE
VR VR+   ID50   CELL   HTC   TI   TI+   LAB PRT CATE	VIA HST VR VR+ DOSE HTC VUH RTE D TOX SP L PR DATE
VR VR+   ID50   CELL   HTC   TI   TI+   LAB PRT CATE	VIA HST VR VR+ DOSE HTC VUH RTE D TOX SP L PR DATE
NOT ACT VERO 32 0 SO HTT 90-03-0  NOT ACT VERO 49 2.92 SO HTT 90-03-0  NOT ACT VERO 43.3 0 SO HTT 90-03-0  NOT ACT VERO 32 0 SO HTT 90-03-0  E NOT ACT VERO 32 0 SO HTT 90-03-0	VIA HST VR VR+ DOSE HTC VCH RTE D TOX SP L PR DATE
NOT ACT   VERO   32   0   SO HTT 90-03-0	VIA HST VR VR+ DOSE HTC VCH RTE D TOX SP L PR DATE
VR VR+   ID50   CELL   HTC   TI   TI+   LAB PRT CATE	VIA HST VR VR+ DOSE HTC VUH RTE D TOX SP L PR DATE
NOT ACT VERO 32 0 SO HTT 90-03-0  NOT ACT VERO 49 2.92 SO HTT 90-03-0  NOT ACT VERO 43.3 0 SO HTT 90-03-0  NOT ACT VERO 32 0 SO HTT 90-03-0  SO HTT 90-03-0  SO HTT 90-03-0	VIA HST VR VR+ DOSE HTC VUH RTE D TOX SP L PR DATE

PLATE U9B

# IN VITRO ANTIVIRAL RESULTS MIT ASSAY

DRUG: AVS 6445 TAI: >17.14 SI: 2.17

_	1	_ 2	3	4	5	6	7	•	9	10	11	12
Γ		i	eagent back	dianue		I			plante backg	round		
A [	0.043	0.042	0.045	0.041	0.040	0.042	0.001	0.001	0.001	0.001	0.001	0.001
Г	T	000/100				ı	104	arug	6446 experie	nenus	00/40	tox
<b>3</b>	- 1	0.918			Į		0.833	0.402	0.373	0.379	0.837	0.871
c	- 1	0.946			1		1.074	0.286	0.430	0.407	0.734	0.870
ם	į	1.094			1		0.830	0.587	0.567	0.534	0.631	0.816
<b>=</b>	- 1	0.295			j		0.882	0.656	0.606	0.590	0.339	0.89
7	ł	0.355			İ	1	0.033	0.036	0.038	0.035	0.369	0.03
a [		0.382					0.035	0.036	0.036	0.036	0.358	0.03
Г									drug <b>6446</b> 00	torime inc bec	aground	
<b>1</b>						ŀ	0.038	0.044	0.043	0.040	0.039	0.039

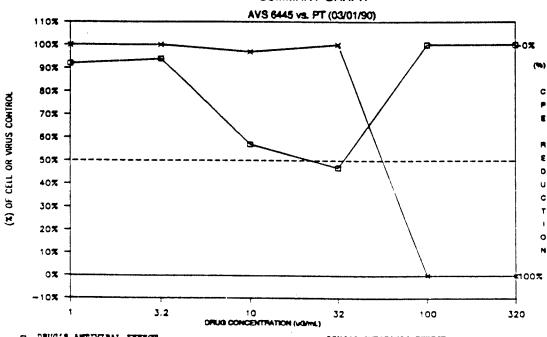
VIRUS CELLS SELPHONT NUMBER STEN	PT VERO 63 ADAMES	Satisfactory: Active: Ret	PROJECT # SPONSOR TEST DATE DATE READ	5975-1 USAMRIID 03/01/90 03/09/90	
REAGENT	0.042	DRUG 6445	25%	50%	95%
VIRUS CONTROL	0.308	TC (uG/mL)	49.00	66.00	96.60
CELL CONTROL	0.811	IC (ug/mL)	5.74	22.60	
DIFFERENTIAL	0.504	ANTIVIRAL INDEX (AI)	0.53	2.92	

	DUNG	6445	ANTIVIRAL 1	TEST VALUES	CYTOTOXICI	TY TEST VALUES	
RO	M OM	COMC.	MEAN	* VIRAL	HEAN	* CKLL	COLORINATALC
21	LATE	(uG/mL)	0.0.	CPS	0.0.	YIABILITY	CONTROL
100	3	1	0.038	924	0.813	100%	003
1	C	3.2	0.028	943	0.933	1004	003
	D	10	0.215	57%	0.703	979	002
	2	32	0.267	479	0.843	100	0.001
	7	100	315	1004	011	0%	0.002
bigi		320	310	100	002	0	004

\* highest drug concentration tested

values shown are final adjusted numbers

#### SUMMARY GRAPH



O DRUG'S ANTIVIRAL EFFECT (% VIRAL CPE)

x DRUG'S CYTOTOXIC EFFECT
(% CELL VIABILITY)

PLATE UAR DRUG 6445

#### IN VITRO ANTIVIRAL RESULTS MTT ASSAY

**DRUG: AVS 6445** TAI: >19.99 SI: 2.53

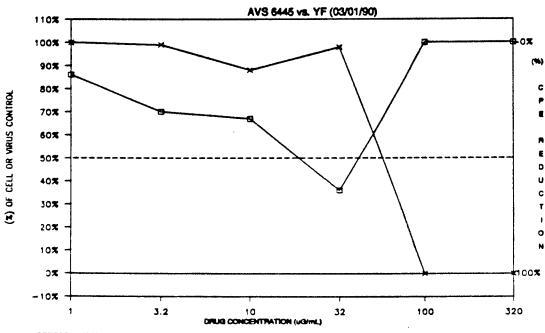
	1	2	3	4	5	6	7	•	•	10	11	12
ſ			reager! back	ground					places barry	mund		
A	0.042	0.040	0.039	0.038	0.039	0.039	0.001	0.001	0.001	0.001	0.001	0.001
- 1	T T	00/146			Ĭ		som	drug	BAAL ORDERIN	merital .	00/148	101
	ŀ	0.847					0.912	0.290	0.356	0.317	0.925	0.943
c		1.048					0.867	0.433	0.430	0.425	0.782	0.925
0		0.922			]		0.767	0.426	0.441	0.474	0.882	0.834
1		0.213			1		0.857	0.621	0.700	0.570	0.218	0.916
7		0.253			I		0.034	0.033	0.034	0.034	0.176	0.042
0	l	0.297			ļ		0.035	0.036	0.035	0.035	0.190	0.034
Г		····		<del></del>					drug dads on	ienmetric bac	kground	
						1	0.038	0.041	0.044	0.039	0.040	0.042

BOLD - highest drug cone

VIRUS CELLS SEIPHENT NUMBER	YF VERO 63	Satisfactory; Active; Ret	est	PROJECT # SPOSSOR TEST DATE	5975-1 USAMRIID 03/01/90
STREE	ASIBI			DATE READ	03/09/90
REAGENT	0.040	DRUG 6445	25%	50%	95%
VIRUS CONTROL	0.185	TC (uC/mL)	48.00	65.30	94.50
CELL CONTROL	0.862	IC (uc/mL)	2.22	18.90	
DIFFERENTIAL	0.677	AFFIVIRAL INCEX (AI)	21.56	3.45	

DR	UG	6445	AMTIVIRAL T	EST VALUES	CYTOTOXICI	TY TEST VALUES		
NOW	0	COMC.	HEAM	♦ VIRAL	KEAN	* CELL	COLORIMETRIC	
PLA	TE	(u0/mL)	O.D.	CPE	O.D.	VIABILITY	CONTROL	
low	3	1	0.094	164	0.865	100	0.003	
	c	3.2	0.204	70%	0.856	991	0.001	
	0	10	0.224	679	0.762	101	001	
		32	0.435	369	0.843	984	0.004	
	,	100	193	100	003	01	0.002	
igh	a -	320	167	100	001	0	002	

### **SUMMARY GRAPH**



DRUG'S ANTIVIRAL EFFECT (% VIRAL CPE)

X DRUG'S CYTOTOXIC EFFECT (% CELL VIABILITY)

### USAMRIID

USAF	IKII	.U		
Antiviral Drug Sc		ning Progra		05/18/90
STRUCTURE	Su	BMITTER 01141.01	CTR NO KN-V-109	AVS NO AVS-006443
· ·	DA	TE RECD 12-28-89	AMT RECEIVED [mg] 86.00	MOL WT (au) 818.979
	HA	ndling/storage		<u> </u>
, D 100 111	so	LUBILITY		
	ST	ABILITY		
	AL	T NAME N3-BENZY	L-2',5'-DI-O-TRITYLU	RIDINE
COMPOUND NAME N3-BENZYL-2',5'-DI-	-0-TR	ITYLURIDINE		
SCREEN INSTRUCTION		IN V	IVO TOXICITY [m	g/kg]
PRIORITY=PT>VEE>YF>KHF>PIC>JE>SF>VV>AD2>VSV		HOST VH RTE LO	SC MTC LAB PR DATE	
IN VITRO SCREEN [ug/ml]	22 01 01 22 01 23 02 22	1	SCREEN [DOSE =	mg/kg]

AMPIVIRAL INDEX: (AI)

_	1	2		4	5	6	7	•	9	10	11	12
ſ			regent beca	Bronne		1			-	round		
A	0.061	0.061	0.061	0.059	0.059	0.062	0.001	0.001	0.002	0.002	0.001	0.002
Γ	tex oc/ve drug 6443 experimental				ton	]		<del></del>		0C/V6		
3	1.421	1.475	0.417	0.536	0.571	1.230	1			1	1.590	
۱ ء	1.514	1.250	0.406	0.411	0.448	0.783	1			1	0.962	
<b>D</b>	1.327	1.605	0.658	0.571	0.499	1.230	- 1			- (	1.398	
	1.281	0.452	0.761	0.771	0.763	1.170	]			ſ	0.414	
P	1.222	0.408	0.872	0.841	0.865	1.158	1			1	0.366	
ေ	0.678	0.466	0.284	0.271	0.382	0.582				ł	0.425	
Γ			drug 6443 con	orimetric bac	#graund					*****	······································	
<b>#</b> [	0.207	0.089	0.068	0.064	0.064	0.064						

VIRUS	YF
CELLS	VERO
SHIPMENT NUMBER	63
STRUI	ASIBI
REAGENT	0.061
VIRUS CONTROL	0.365
CELL CONTROL	1.321
DIFFERENTIAL	0.956

			PROJECT /	5975-1		
3	Satisfactory: Active: Retest SPONSOR USAMRID					
	TOXICITY REBUM		TEST DATE	ATE 03/22/90		
			DATE READ	03/30/90		
٢	DRUG 6443	25%	50%	95%		
Γ	TC (uG/mL)	427.00	750.00	> 1000.00		
ı	20 1.0/025	1	1			

7.54

DRUG	6443	ANTIVIRAL T	EST VALUES	CALOLOXICI	TY TEST VALUES	
NOW ON	CONC.	MEAN	% VIRAL	HOLAN	* CELL	COLORIMETRIC
PLATE	(ug/mt)	0.0.	CPE	0.0.	VIABILITY	CONTROL
O 1	3.2	0.079	92	1.261	95	0.004
c	10	000	100	1.084	82%	0.004
D	32	0.147	05 <b>%</b>	1.214	924	0.004
1	100	0.332	65%	1.157	004	0.008
7	320	0.405	584	1.101	83%	0.029
sigh d	1000	260	100	0.423	329	0.147

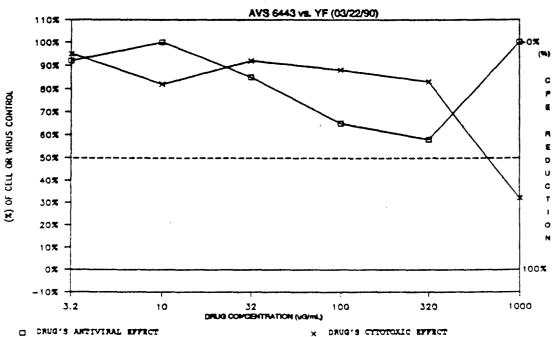
\* highest drug concentration tested

enedmun beteujbs lanit era meone seular

0.00

0.00

#### SUMMARY GRAPH



ORUG'S ANTIVIRAL EFFECT
(% VIRAL CPE)

(# CELT AIMBILITA)

× DERGAR CALCLOYIC EAASCE

SOUTHERN RESEARCH INSTITUTE

US	AMRIII			
Antiviral Drug S	CHIE	ing Progra	CTR NO	05/19/90 AVS NO
STRUCTURE CHIRA	AL SUE	01141.01	KN-VII-83	AVS-006444
	DAT	E RECD	AMT RECEIVED [mg]	MOL WT (au)
°		12-28-89	79.00	726.837
	HAN	DLING/STORAGE		<u> </u>
NH				
, , , , , , , , , , , , , , , , , , ,				
	sor	LUBILITY		
	STA	ABILITY		
	-			
	ALI	NAME 3'-DEOXY-2'	,5'-DI-O-TRITYL-3'-0	XOURIDINE
COMPOUND NAME				
3'-DEOXY-2',5'-DI-0	TRITYL-	-3'-OXOURIDINE	:	
SCREEN INSTRUCTION		TN V	'IVO TOXICITY (n	ng/kg]
PRIORITY=PT>VEE>YF>KHF>PIC>JE>SF>VV>AD2>VSV			DSO MTC LAB PR DATE	.97 .49 )
T. V.		7031 74 812 2	270 200 10 2010	
IN VITRO SCREEN [ug/ml]		TN 1/71/	O SCREEN (Dose	mg/kg]
·				
VIR         VR         VR+         1050         CELL         HTC         TI         TI+         LAB PRT DAT           HIV         NOT ACT         HTZ         > 100         0         SO HTZ 90-           JE         NOT ACT         VERO         51         0         SO HTT 90-	-03-20	VIR HST VR VR+	DOSE MIC VEH RIE D TOX S	SP L PR DATE
JE NOT ACT VERO 547 0 SO HTT 90- PT 79.2 VERO > 320 > 4.04 SO HTT 90-	-03-22			
PT NOT ACT VERO 257 0 SO MTT 95 NOT ACT VERO 30 0 SO MTT 95 F NOT ACT VERO 365 0 SO MTT 90 NOT				
VEE NOT ACT VERO 680 0 SO MTT 90- VEE NOT ACT VERO > 120 0 SO MTT 90-	-03-23			
VV NOT ACT VERO 116 0 SO HTT 90- YF 29.0 VERO 410 24.55 SO HTT 90- YF MOT ACT VERO > 320 0 SO HTT 90-	-03-22			
YF NOT ACT VERG > 320 0 SO MTT 90-	-03-01			
1		1	•	

PLATE UQO

# IN VITRO ANTIVIRAL RESULTS MTT ASSAY

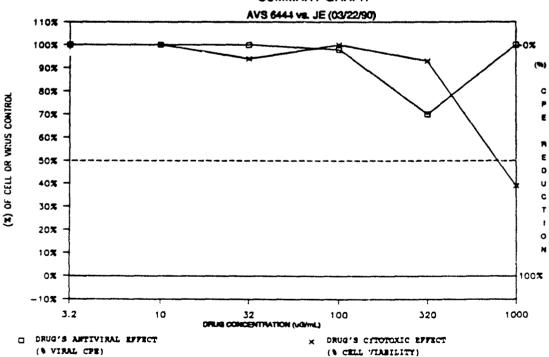
DRUG: AVS 6444
TAI: 3.21 SI: ----

	1	2	3	4	5	6	7		9	10	11	12
Г			rengent beck	Stanue					plante backg	round		
A	0.077	0.073	0.071	0.070	0.072	0.072	0.001	0.001	0.002	0.001	0.002	0.002
Г		000/40			i		los	drug	8444 experie	nental .	00/40	tox
	- 1	1.519			ŀ		1.517	0.509	0.525	0.554	1.544	1.671
c	1	1.476			1		1.463	0.458	0.481	0.478	1.484	1.577
٥		1.487					1.474	0.462	0.467	0.492	1.537	1.384
	ſ	0.607			ļ		1.542	0.621	0.651	0.540	0.589	1.615
7		0.615			l		1.327	0.845	0.897	0.927	0.619	1.517
a l		0.626			Ì		0.697	0.474	0.515	0.524	0.613	0.680
Γ									trug 8444 pont	ionmetric bac	kground	
							0.121	0.082	0.081	0.078	0.078	0.077

VIRUS CELLS SEIPHERT HUNDER STRE	JE VERO 63 HARAYAHI	Satisfactory; Active; Ret TOXICITY REBUS	<u>••ξ</u>	PROJECT # SPONSOR TEST DATE DATE READ	5975-1 USAMRIID 03/22/90 03/29/90
REAGENT	0.073	DRUG 6444	25%	50%	95%
VIRUS CONTROL	0.539	TC (uG/mL)	547.00	861.00	> 1000.00
CELL CONTROL	1.436	IC (nd/ml)	260.00		
DIFFERENTIAL	0.897	ANTIVINAL INDEX (AI)	2.10		

DRUG 5444		5444	ANTIVIRAL	TEST VALUES	CYTOTOXICI		
NOW	CS	CONC.	HEAM	VIRAL	MEAN	* CELL	COLORIMETRIC
PL	ATE	(uG/mL)	O.D.	CPE	O.D.	VIABILITY	CONTROL
104		3.2	087	100%	1.517	100	0.005
	C	10	145	1000	1.452	100	0.006
	D	32	144	100	1.351	941	0.006
		100	0.017	947	1.497	100	0.009
	7	320	0.268	709	1.340	939	0.010
high		1000	156	100	0.567	39%	0.049

#### **SUMMARY GRAPH**



32

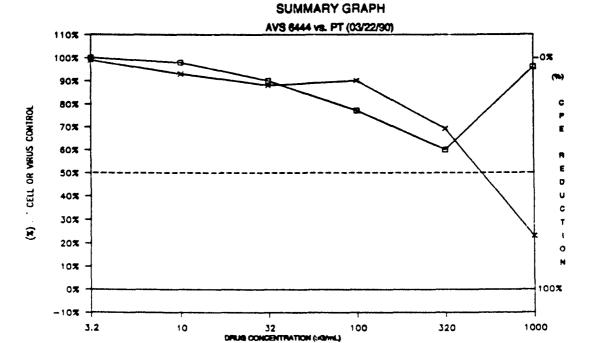
	1	2	3	4	5	•	7		9	10	11	12
٦			reagent back	Bround					plastic backg	round		
١I	0.054	0.055	0.054	0.051	0.051	0.053	0.002	0.002	0.001	0.001	0.001	0.00
Г		00/14					1que	drug	6444 experim	nen tal	cove	lox
•	1	1.408			l		1.654	0.253	0.349	0.342	1.305	1.30
:	1	1.631			1		1.553	0.377	0.414	0.423	1.539	1.23
o	Ĺ	1.634			1		1.452	0.477	0.492	0.519	1.485	1.20
	ſ	0.388					1.415	0.593	0.743	0.580	0.418	1.29
,	1	0.391			- 1		1.118	0.988	0.750	0.778	0.386	1.00
١	-	0.316			- 1		0.384	0.422	0.430	0.447	0.403	0.38
Г									drug 6444 00	iorimetric bac	kground	
<b>a</b> [							0.055	0.057	0.056	0.055	0.051	0.04

VIRUS CELLS SETMENT HUMBER STREE	PT VERO 63 ADAMES	Satisfactory: Active: Ret TOXICITY REPUR	<b>.</b>	PROJECT # SPONSOR TEST DATE DATE READ	5975-1 USANTID 03/22/90 03/30/90
REAGENT	0.053	DRUG 6444	25%	50%	95%
VIRUS CONTROL	0.331	TC (wG/mL)	257.00	601.00	> 1000.00
CELL CONTROL	1.447	IC (uc/al)	115.00		
DIFFERENTIAL	1.117	APTIVIRAL INDEX (AI)	2.24		

Q	RUG	6444	ANTIVIRAL 1	TEST VALUES	CALOLOXICI:	TY TEST VALUES	
ROM	ON	COMC.	HEAD	• VIRAL	KELAH	* CELL	COLORIMETRIC
PL	ATE	(uG/mL)	0.0.	CPE	O.D.	VIABILITY	CONTROL
104	3	3.2	065	100	1.429	994	004
	c	10	0.023	204	1.345	939	002
	D	32	0.110	90%	1.276	867	0.002
	1	100	0.252	779	1.300	909	0.003
	7	320	0.451	60%	1.003	699	0.004
high		1000	0.047	964	0.327	231	0.002

\* highest drug concentration teste

values shown are final adjusted numbers



DRUG'S AMTIVIRAL EFFECT (% VIRAL CPE) DRUG'S CYTOTOXIC EFFECT (% CELL VIABILITY)

	USAMRI	ID		
Antinian 1 D	Camaa	ning Progra	m	
Antiviral Dr STRUCTURE	CHIRAL SI	UBMITTER	CTR NO	05/18/90 AVS NO
	_	01141.01	KN-VII-21	AVS-006445
	D	ATE RECD 12-28-89	AMT RECEIVED [mg] 74.00	MOL WT (au) 726.837
	Н.	ANDLING/STORAGE		
NH	""	AND ELING, STORAGE		
	S	OLUBILITY		
"				
	s <sup>,</sup>	TABILITY		
	1	LT NAME		
·	6		5'-DI-O-TRITYL-2'-0	XOURIDINE
COMPOUND NAME				
2'-DEOXY-3',5'-	-DI-O-TRITY	L-2'-OXOURIDINE		
SCREEN INSTRUCTION		IN V	IVO TOXICITY [m	g/kg]
PRIORITY=PT>VEE>YF>KHF>PIC>JE>SF>VV>AD2>VSV		HOST VH RTE LO	SO HTC LAS PR DATE	
IN VITRO SCREEN [ug/ml]		IN VIVO	SCREEN [Dose =	mg/kg]
	RT DATE	VIR HST VM VR+ D	CSE MTO VEH RIE D TOX S	P L PR DATE
JE NOT ACT VERO 49 0 SO H	TT 90-03-20 TT 90-03-22 T: 90-03-01			
PT 22.6 VERO 49 2.92 SO F PT MOT ACT VERO 8.87 0 30 H	TT 90-03-01 TT 90-03-22			
SF NOT ACT VERO 43.3 0 SO H	TT 90-01-22 TT 90-03-01 TT 90-03-02	j		
VEE         NOT ACT         VERO         44         0         50 H           VV         HOT ACT         VERO         4.6         0         50 H	TT 90-03-23 TT 90-03-22			
	TT 90-03-01 TT 90-03-22			
		1		

### PLATE UQA DRUG 6445

# IN VITRO ANTIVIRAL RESULTS MTT ASSAY

AMPIVIRAL INDEX (AI)

DRUG: AVS 6445 TAI: >18.33 SI: 3.27

	1	2	3	4	5	6	7		9	10	11	12
Г			reagent bace	ground					pleases backg	round		
A	0.063	0.063	0.062	0.060	0.056	0.061	0.001	0.001	0.001	0.001	0.001	0.001
Г	lest .	00/10	drug	6445 experim	TON LOS	1Q.E					00/ve	
<b>3</b>	1.549	1.409	0.619	0.596	0.557	1.504	1			i	1.418	
c	1.442	1.368	0.740	0.005	0.674	1.607				1	1.192	
D	1.368	1.448	0.998	0.971	0.924	1.488	i			į	1.550	
8	1.067	0.526	0.934	0.600	0.848	1.022				[	0.433	
7	0.062	0.481	0.051	0.050	0.054	0.051	٠				0.380	
9	0.066	0.494	0.066	0.061	0.059	0.065				i	0.400	
Г			drug BLAS oo	ioremetris bec	kground							
	0.075	0.071	0.065	0.062	0.064	0.062						
-	ton-ood to	merly oo-	sed son troi	40-4M/16 00	red	900	- highest dr.	46 00ne		values sho	NAME OF THE PARTY NAME OF THE	denertee

VIRUS	YF
CELLS	VERO
SEIPHENT NUMBER	63
STRE	AS ISI
REAGENT	0.061
VIRUS CONTROL	0.392
CELL CONTROL	1.370
DIFFERENTIAL	0.979

atisfactory; Active; R	etest	PROJECT # SPONSOR	5975-1 USAHRIID
POXICITY RERUN		TEST DATE	03/22/90
		DATE READ	03/30/90
DRUG 6445	25%	50%	95%
TC (ug/mL)	29.60	52.80	95.30
IC (uc/ml)	2.35	9.06	

12.63

DNUG		6445	45 AMTIVIRAL TEST VALUES		CYTOTOXICI		
ROW	OM	CONC.	HOLAN	% VIRAL	HEAM	♦ CELL	COLORINGTRIC
PLA	TE	(ug/mL)	O.D.	CPE	0.0.	VILBILITY	CONTROL
low		1	0.137	467	1.465	1000	0.001
	c	3.2	0.284	719	1.461	100	0.003
	D	10	0.511	487	1.366	100	0.001
	1	32	0.338	65%	0.980	721	0.004
	7	100	411	100	014	04	0.010
high		320	404	100	009	0	0.014

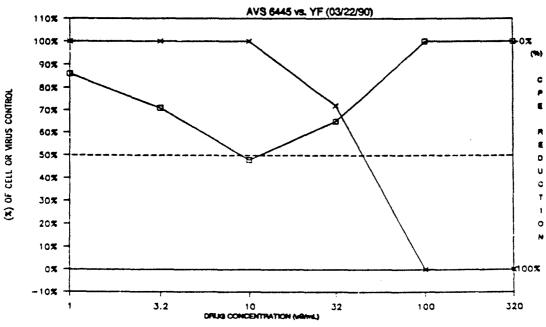
\* highest drug concentration tester

values shown are final adjusted numbers

5.83

0.00

### SUMMARY GRAPH



DRUG'S AMTIVIRAL EFFECT (% VIRAL CPE)

DRUG'S CYTOTOXIC EFFECT (% CELL VIABILITY)

				Antiviral	Drug Scre	ening Progra	ım	05/18/90
TRUCTU	RE				CHIRAL	SUBMITTER 01141.01	CTR NO KN-II-53	AVS NO AVS-006449
				0		DATE RECD 12-28-89	AMT RECEIVED [mg] 75.00	MOL WT (au) 284.271
					}	HANDLING/STORAGE		
				NH L				
		<u>~</u>	·	N O	}	SOLUBILITY		
	• H	0			Ì			
				)	ļ.	STABILITY		
			$\times$					
						ALT NAME	-O-ISOPROPYLIDINEUR	IDINE
MPOUND !	NAME			······································				
				2',3'	-O-ISOPROPYLI	DINEURIDINE		
				TRUCTION		IN V	IVO TOXICITY [	mg/kg]
iority=	PT>VEE>Y	F>KHF>	PIC>JE>S	F>VV>AD2>VSV		HOST VH RTE LO	SO MTC LAS PR DATE	
	IN	VITR	O SCREE	N [ug/ml]		IN VIVO	SCREEN [Dose	mg/kg]
VR VI	NOT ACT	CELL MT2	MTC > 100	TI TI+	SO HTT 90-03-20	VER HST VR VR+ C	NOSE MIC VEH RIE D TOX	SP L PR DATE
	NOT ACT NOT ACT 264	VERO VERO	466 > 320 > 320	0 0 > 1.21	50 HTT 90-03-22 50 HTT 90-03-01 50 HTT 90-03-01			
	NOT ACT NOT ACT	VERO VERO	363 472 > 320	0	SO MTT 90-03-22 SO MTT 90-03-22 SO MTT 90-03-01			
	NOT ACT NOT ACT NOT ACT	VERO VERO VERO	251 320 195 517	0 0 0	SO MTT 90-03-02 SO MTT 90-03-23 SU MTT SO MTT 90-03-22			
	NOT ACT	VENO	> 320		SO HTT 90-03-01			
		·						

				USAMR	IID		
		Ą	ntivir	al Drug Scre	ening Program		25/:
STRUCTURE					SUBMITTER 01141.01	CTR NO MS-I-47	AVS NO AVS-C06456
					DATE RECD 12-28-89	AMT RECEIVED [mg] 79.20	MOL WT (au) -544.694
		$\rightarrow$	\ \	<b>&gt;</b>	HANDLING/STORAGE		
•	) , <	<b>***</b>	✓	,	SOLUBILITY		****
Na <sup>+</sup> o"					STABILITY		
					ALT NAME SODIUM ETHYL(CHO	LESTERYLOXYCARBONYL)	-PHOSPHONATE
CMPOUND NAME		SOD	ICM ETHYL	(CHOLESTERYLOXYO	ARBONYL) -PHCSPHON	ATE	
	SCREE	N INSTE	RUCTION		IN V	IVO TOXICITY [n	ng/kg]
RIORITY-PT>VE	E>YF>KHF>P	'IC>JE>SF	>VV>AD2>\	'sv	HOST VH RTE LD	50 MTC LAB PR DATE	
	IN VITRO	SCREEN	l (ug/r	nl]	IN VIVO	SCREEN [Dose	mg/kg]
IV NOT		HTC 49	<u>71 T</u>	SO HTT	VIR HST VR VR+ D	OSE MTC VEH ATE D TOX S	SP L PR DATE
E NOT	ACT VERO	25.6 47.4	0	SO NTT SO NTT 90-03-06			
T NOT T NOT F NOT	ACT VERO	41.9 43.3 56.7	0 0	SO HTT 90-03-06 SO HTT SO HTT 90-03-06			
F NOT	ACT VERO	72.5 116	0	SO HTT SO HTT			
EE NOT V NOT	ACT VERO	54.7 21.8	0	SO MTT 90-03-09 SO MTT	į		
F NOT	61 VERO ACT VERO	132 40.3	3.25	SO HTT SO HTT 90-03-06			
					i		

PLATE UDN

# IN VITRO ANTIVIRAL RESULTS MTT ASSAY

DRUG: AVS 6456 TAI: 3.19 SI: ----

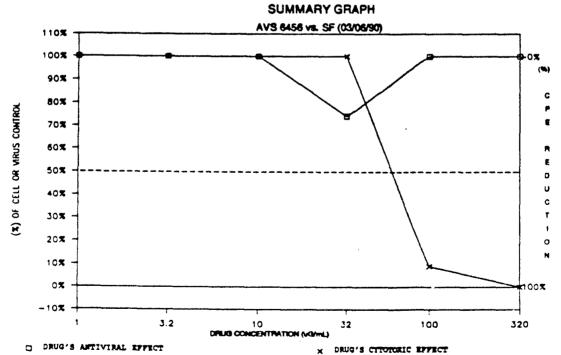
_	1	2	3	4	5	6	7		•	10	11	12
			reagent back	tground					piants barry	ound		
a [	0.059	0.057	0.063	0.055	0.063	0.054	0.001	0.001	0.002	0.002	0.001	0.002
Γ	100	00/10	gunb	6466 experie	Pental	101					00/46	
•	0.936	0.768	0.225	0.228	0.213	0.841				1	0.941	
c	0.893	0.819	0.172	0.162	0.145	0.951				- 1	1.024	
١٩	0.969	0.750	0.144	0.136	0.135	1.052				1	0.789	
	0.964	0.281	0.432	0.408	0.423	0.955				Ī	0.268	
7	0.174	0.271	0.053	0.051	0.052	0.077	ĺ			ĺ	0.265	
ے ا	0.050	0.260	0.046	0.044	0.044	0.043					0.283	
Γ			drug 6466 oo	present per	#ground					<u></u>		
<b>8</b> [	0.050	0.053	0.055	0.057	0.052	0.055						
	100-444 14	micity co-	ion mon bea	ACMARINE COL	wat	BOLD	- highest dru	4 0004		values ofto	Wn 2/0 00001	densities

VIRUS CELLS SHIPHENT HUGGER STRII	SF VERO 63 SICILIA	Setisfactory; Active; Net	PROJECT # SPORSOR TREE DATE DATE READ	5975-1 UBANRIID 03/06/90 03/14/90	
REACENT	0.059	DRUG 6456	25%	50%	95%
VIRUS CONTROL	0.216	TC (u9/mL)	50.70	69.40	190.00
CELL CONTROL	0.790	IC (ug/mL)	10.60		
DIFFERENTIAL	0.574	AMPINIRAL INDEX (AT)	1.66		

DRUG 6456		6456	ANTIVIRAL T	EST VALUES	CITOTOXICI	<del></del>	
ROW	OM	CONC.	HEAR	• VIRAL	MRAN	* CELL	COLORINETRIC
P1,	ATE	(uG/mL)	0.0.	CPE	0.5.	VIABILITY	COFTROL
low	3	1	049	100	0.834	100	004
	C	3.2	108	100	0.871	100	007
	D	10	135	100%	0.963	100	001
	2	32	0.150	749	0.905	100%	004
	7	100	217	1000	0.073	97	006
high	a -	320	222	100	004	01	000

\* highest drug concentration sessed

values shown are final adjusted numbers



( CELL VIABILITY)

(% VIRAL CPE)

### DITION IN VIVO TOXICITY [mg/kg]  IN VITRO SCREEN [ug/ml]  IN VIVO SCREEN [Dose = mg/kg]  VI VIV. 1053 CELL NTC TI TI: LAB PRT DATE   VIR PST VIR VIR DOSE NTC VER ATE D TOX SP L PR DATE   NOT ACT VERD > 1000 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0		USAMRII	.D			
SUBMITTER  SUBMITTER  OATE RECD  ATT RECD   AMT RECEIVED (mg)   MOL NT (au)  634.329  HANDLING/SIGRAGE  SOLUBILITY  ALT NAME  DI-ISOBUTYL(CHOLESTERYLOXYCARBONYL)-PHOSPHONATE  OTHER SCREEN INSTRUCTION   IN VIVO TOXICITY [mg/kg]  SCREEN INSTRUCTION   IN VIVO TOXICITY [mg/kg]  IN VITRO SCREEN [ug/ml]   IN VIVO SCREEN [Dose = mg/kg]  VALUE   1035   CELL   NTC   11   144 PRT DATE   VIR NOT ACT	Antiviral D	rua Scree	ning Progra	ım	03/18/	
12-28-89   70.40   634.929	TRUCTURE		BMITTER	CTR NO	AVS NO	
SOLUBILITY  ALT NAME DI-ISOBUTYL (CHOLESTERYLOXYCARBONYL) -PHOSPHONATE  SCREEN INSTRUCTION  IN VIVO TOXICITY [mg/kg]  IN VITRO SCREEN [ug/ml]  IN VIVO TOXICITY [mg/kg]  VN VN 1010 CLL NT 71 (1. LOS PRT DATE  VN VN 1010 CLL NT 71 (1. LOS PRT DATE  VN VN VN 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1. LOS PRT DATE  VN N N 1010 CLL NT 71 (1.		DA			1	
MFOUND NAME  DI-ISOBUTYL (CHOLESTERYLOXYCARBONYL) - PHOSPHONATE  SCREEN INSTRUCTION  IN VIVO TOXICITY [mg/kg]  LORITY-PT>VEE>YF>KHF>PIC>JE>SF>VV>AD2>VSV  IN VITRO SCREEN [ug/ml]  IN VIVO SCREEN [Dose = mg/kg]  VN VN 1050 CELL MTC II II IAB PRI DATE  WOT ACT WIRD 3120 0 50 MTT  WOT ACT VERD 1320 0 50 MTT 10-03-04  MCT ACT VERD 1320 0 50 MTT 10-03-04		НА	NDLING/STORAGE		L	
ALT NAME DI-ISOBUTYL(CHOLESTERYLOXYCARBONYL)-PHOSPHONATE  SCREEN INSTRUCTION  IN VIVO TOXICITY [mg/kg]  IORITY-PT>VEE>YF>KHF>PIC>JE>SF>VV>AD2>VSV  IN VITRO SCREEN [ug/ml]  IN VIVO SCREEN [DSG MTC LAB PR DATE  IN VIVO SCREEN [DGG = mg/kg]  VR VR 1050 CELL MTC TI 1: LAB PRT DATE  WOT ACT MT2 > 100 0 SO MTT M0-03-04  MOT ACT WEND > 120 0 SO MTT M0-03-04  MOT ACT VEND > 120 0 SO MTT M0-03-04  MOT ACT VEND > 120 0 SO MTT M0-03-04  MOT ACT VEND > 120 0 SO MTT M0-03-04  MOT ACT VEND > 120 0 SO MTT M0-03-04  MOT ACT VEND > 120 0 SO MTT M0-03-04  MOT ACT VEND > 120 0 SO MTT M0-03-04  MOT ACT VEND > 120 0 SO MTT M0-03-04  MOT ACT VEND > 120 0 SO MTT M0-03-04  MOT ACT VEND > 120 0 SO MTT M0-03-04  MOT ACT VEND > 120 0 SO MTT M0-03-04  MOT ACT VEND > 120 0 SO MTT M0-03-05  MOT ACT VEND > 120 0 SO MTT M0-03-05  MOT ACT VEND > 120 0 SO MTT M0-03-05  MOT ACT VEND > 120 0 SO MTT M0-03-06  MOT ACT VEND > 120 0 SO MTT M0-03-06		so	DEUBILITY			
DI-ISOBUTYL (CHOLESTERYLOXYCARBONYL) - PHOSPHONATE  SCREEN INSTRUCTION  IN VIVO TOXICITY [mg/kg]  IN VITO SCREEN [ug/ml]  IN VIVO SCREEN [Dose = mg/kg]  VN VN 1050 CELL NTC TI T: LAB PRI DATE  VN NT ACT NT2 > 100 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06  NOT ACT VN > 320 0 50 NTT 100-30-06	<b>→</b>	ST	ABILITY			
SCREEN INSTRUCTION				LESTERYLOXYCARBONYL)	-PHOSPHONATE	
IN VITRO SCREEN [ug/ml]  IN VITRO SCREEN [ug/ml]  VR VR* 1050 CELL MTC II II* LAB PRI DATE  VIR MST VR VR* 0050 CELL MTC II II* LAB PRI DATE  ( NOT ACT MT2 > 100 0 50 MTT 90-03-06  NOT ACT WEND > 320 0 50 MTT 90-03-06  NOT ACT VEND > 320 0 50 MTT 90-03-06  NOT ACT VEND > 320 0 50 MTT 90-03-06  NOT ACT VEND > 320 0 50 MTT 90-03-06  NOT ACT VEND > 320 0 50 MTT 90-03-06  NOT ACT VEND > 320 0 50 MTT 90-03-06  NOT ACT VEND > 320 0 50 MTT 90-03-06  NOT ACT VEND > 320 0 50 MTT 90-03-08  NOT ACT VEND > 320 0 50 MTT 90-03-08  NOT ACT VEND > 320 0 50 MTT 90-03-08  NOT ACT VEND > 320 0 50 MTT 90-03-08  NOT ACT VEND > 320 0 50 MTT 90-03-06  NOT ACT VEND > 320 0 50 MTT 90-03-06	DMPOUND NAME  OI-ISOBUTYL(CHOLE	STERYLOXYCARE	ONYL) -PHOSPHON	ATE		
IN VITRO SCREEN [ug/ml]  IN VIVO SCREEN [Dose = mg/kg]  VR VR- 1050 CELL HTC TI TI- LAB PRT DATE  NOT ACT HT2 > 100 0 SO HTT NOT ACT VERO > 120 0 SO HTT 90-03-06 NOT ACT VERO > 120 0 SO HTT 90-03-06 NOT ACT VERO > 120 0 SO HTT 90-03-06 NOT ACT VERO > 120 0 SO HTT 90-03-06 NOT ACT VERO > 120 0 SO HTT 90-03-06 NOT ACT VERO > 120 0 SO HTT 90-03-06 NOT ACT VERO > 120 0 SO HTT 90-03-06 NOT ACT VERO > 1000 0 SO HTT 90-03-09 SO HTT 90-03-09 SO HTT 90-03-09 HOT ACT VERO > 120 0 SO HTT 90-03-03 NOT ACT VERO > 120 0 SO HTT 90-03-03 NOT ACT VERO > 120 0 SO HTT 90-03-03 NOT ACT VERO > 120 0 SO HTT 90-03-03 NOT ACT VERO > 120 0 SO HTT 90-03-04 NOT ACT VERO > 120 0 SO HTT 90-03-04	SCREEN INSTRUCTION		IN V	IVO TOXICITY [m	g/kg]	
VR VR*   1050   CELL   HTC   TI   TI*   LAB PRT DATE   VIR HST VR VR*   DOSE   MTC   VEH RTE D TOX SP L PR DATE	RIORITY=PT>VEE>YF>KHF>PIC>JE>SF>VV>AD2>VSV		HOST VM RTE LDS0 MTC LAB PR DATE			
NOT ACT   HT2   100   0   SO   HTT   HOT ACT   VENO   320   0   SO   HTT   90-03-06   HOT ACT   VENO   2000   0   SO   HTT   90-03-22   HOT ACT   VENO   320   0   SO   HTT   90-03-26   HOT ACT   VENO   320   0   SO   HTT   90-03-26   HOT ACT   VENO   320   0   SO   HTT   90-03-06   HOT ACT   VENO   1000   0   SO   HTT   90-03-22   HOT ACT   VENO   1000   0   SO   HTT   90-03-22   HOT ACT   VENO   1000   0   SO   HTT   90-03-23   HOT ACT   VENO   320   0   SO   HTT   90-03-23   HOT ACT   VENO   320   0   SO   HTT   90-03-06   HOT ACT   POOL ACT	IN VITRO SCREEN [ug/ml]		IN VIVO	) SCREEN [Dose =	mg/kg]	
NOT ACT VERO > 1000 0 SO MTT 90-03-22 NOT ACT VERO > 1000 0 SO MTT 90-03-06 NOT ACT VERO > 1000 0 SO MTT 90-03-22 NOT ACT VERO > 1000 0 SO MTT 90-03-06 NOT ACT VERO > 1000 0 SO MTT 90-03-08 NOT ACT VERO 155 0 SO MTT 90-03-09 NOT ACT VERO > 1000 0 SO MTT 90-03-23 NOT ACT VERO > 1000 0 SO MTT 90-03-09 NOT ACT VERO > 1000 0 SO MTT 90-03-06 NOT ACT VERO > 120 0 SO MTT NOT ACT VERO > 120 0 SO MTT NOT ACT VERO > 120 0 SO MTT NOT ACT VERO > 120 0 SO MTT NOT ACT VERO > 120 0 SO MTT	V NOT ACT MT2 > 100 0 SC	HTT	VIR AST VR VR+	SOSE MIC VEH RIE D TOX S	P L PR DATE	
	NOT ACT	0 MTT 90-03-22 0 MTT 90-03-06 0 MTT 90-03-22 0 MTT 90-03-22 0 MTT 90-03-23 0 MTT 90-03-23 0 MTT 90-03-23 0 MTT 90-03-23				

PLAT	E UC
DRUG	6458

### IN VITRO ANTIVIRAL RESULTS MTT ASSAY

**DRUG: AVS 6458** TAI: >0.50 SI: ---

	1	2	3	4	5	6	7		9	10	11	12
٦			reagent back	ground					piere backg	round		
A	0.062	0.059	0.057	0.058	0.057	0.058	0.002	0.001	8.001	0.002	0.001	0.001
Г	100	00/10	drug	6468 experim	nental	lost	I				00/10	
3	1.334	1.245	0.411	0.439	0.415	1.113	i				1.215	
c	1.259	1.243	0.405	0.394	0.392	1.110				1	1.185	
٥	1.290	1.203	0.365	0.422	0.412	1.110				1	1.211	
2	1.203	0.413	0.454	0.375	0.451	1.054	1			ſ	0.442	
7	1.226	0.403	0.443	0.430	0.497	1.030				1	0.455	
a	1.024	0.393	0.689	0.669	0.689	0.862					0.446	
Γ			Srug 8468 00	iorimetric bur	kground			·				
=	0.058	0.061	0.061	0.060	0.060	0.061						

VIRUS	PT
CELLS	VERO
SHIPMENT NUMBER	63
STRE	ADAMES
REAGENT	0.059
VIRUS CONTROL	0.367
CELL CONTROL	1.172

0.805

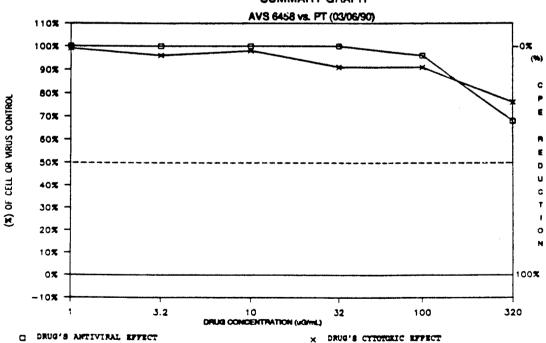
DIFFERENTIAL

		PROJECT #	5975-1
Satisfactory: Active: R	SPORSOR	USAKRIID	
		TEST DATE	03/06/90
		DATE READ	02/16/90
DRUG 6458	25%	50%	95%

DRUG 6458	25%	50%	95%
TC (uG/mL)	> 320.00	> 320.00	> 320.00
IC (ue/mL)	239.00		
ANTIVIRAL INDEX (AI)	> 1.34		

DRUG 6458		6458	ANTIVIRAL T	EST VALUES	CYTOTOXICI			
ROW	OM	COMC.	HORAM	* VIRAL	KEAN	* CELL	COLORIMETRIC	
PL	ATE	(ug/mL)	O.D.	CPE	0.0.	VIABILITY	CONTROL	
low	3	1	006	1004	1.163	999	0.002	
	C	3.2	029	100	1.125	961	0.001	
	D	10	027	100	1-145	981	0.001	
	E	32	001	100	1.068	919	0.002	
	7	100	0.029	96%	1.068	919	0.002	
iigh		320	0.257	669	0.865	76	0.000	

#### **SUMMARY GRAPH**



X DRUG'S CYTOTORIC EFFECT (\* CELL VINBILITY)

Antiviral Drug S	AMRIII			***************************************	
	Screen	ina Progra	am.		
RUCTURE CHIRA	200	MITTER	CTR NO	05/18/9 AVS NO	
		01141.01	KN-II-71	AVS-006462	
	DAT	E RECD	AMT RECEIVED [mg]		
HC1 NH <sub>2</sub>		12-28-89	72.40	325.729	
N	HAN	DLING/STORAGE			
	]				
N O O					
но	SOI	UBILITY			
0,0	STA	BILITY			
S II					
٥					
	ALI	NAME	LFINYLCYTIDINE HYDRO	CHIODIDE	
		2 , 3 -0-30	LFINIECTITOTAE NIDRO	CHECKIDE	
MPOUND NAME		V850001100105			
2',3'-0-SULFINYLCY	IIDINE P	IDROCHECKIDE			
SCREEN INSTRUCTION		IN VIVO TOXICITY [mg/kg]			
IORITY-PT>VEE>YF>KHF>PIC>JE>SF>VV>AD2>VSV		HOST VH RTE L	050 MTC LAB PR DATE		
IN VITRO SCREEN [ug/ml]	Σ		O SCREEN [Dose =	mg/kg]	
NOT ACT MT2 .06 0 SO MTT NOT ACT MT2 < .32 0 SO MTT NOT ACT VERO 22.4 0 SO MTT 90=	01-04				
NOT ACT VERO 36.6 0 50 HTT 90-1 NOT ACT VERO 21 0 50 HTT 90-1	-03-06				
NOT ACT VERO 9.73 0 SO MTT 90-4 1.72 VERO 16.7 14.12 SO MTT					
3.28 VERO 25.7 18.66 SO HTT HOT ACT VERO 44.4 0 SO HTT 90-1	-03-06				
	'				

PLATE OSS

# IN VITRO ANTIVIRAL RESULTS MTT ASSAY

DRUG: AVS 6462 TAI: >32.30 SI: 7.85

1	2	3	4	5	6	7	•	9	10	11	12
		eagent back	ground					pleases beening	round		
0.104	0.114	0.112	0.111	0.111	0.112	0.000	0.000	0.000	0.000	0.900	0.000
	09/19					X/E	drug	8462 experts	nental	oc/ve	loz
	1.542			1		1.492	0.362	0.278	0.427	1.506	1.619
- 1	1.596					1.491	0.820	0.842	0.963	1.508	1.676
į	1.651			1		1.650	1.412	1.560	1.551	1.524	1,770
	0.160			1		1.040	0.929	0.938	0.452	0.247	1.056
- 1	0.248			}		0.573	0.511	0.523	0.524	0.174	0.542
	0.213			Į		0.320	0.312	0.315	0.305	0.238	0.349
								drug \$462 00	torimetrie bac	kground	
						0.153	0.123	0.110	0.108	0.110	0.117
105-010 10	autotty co-c	ed control	vo-virue cor	roi	900	- highest dru	e cone		Values end	wn are optica	deneruer

•	VIRUS	W
	CELLS	VERO
	SELFIGHT FUGER	63
	STREE	LEDCA
	REAGENT	0.111
	VIRUS CONTROL	0.103
	CELL CONTROL	1.444
	DIFFERENTIAL	1.341

	PROJECT #	5975-4
Satisfactory; Active; Retest	SPORSOR	USAMMIID
RETEST AT 100 UG/ML	TEST DATE	03/29/90
	DATE READ	04/04/90

DRUG 6462	25%	50%	95%
TC (uG/mL)	25.70	61.10	> 320.00
IC (uG/mL)	1.56	3.26	9.54
ANTIVIRAL INDEX (AI)	16.44	18.66	> 33.56

DRUG 6462			ANTIVIRAL 1	TEST VALUES	CYTOTOXICI	TY TEST VALUES	
ROM	9	COMC.	XEAR	• VIRAL	HEAM	9 CELL	COLORIMITRIC
PL	ATE	(ug/ml)	0.0.	CPE	0.0.	VIABILITY	CONTROL
104	3	1	0.136	90%	1.439	130	0.006
	c	3.2	0.563	51%	1.474	1004	001
	0	10	1.297	3%	1.602	100%	003
		32	0.694	489	0.939	65%	001
	7	100	0.294	789	0.435	30%	0.012
1gh	9 4	320	0.055	96થ	0.102	134	0.042

\* highest drug gangentragen tester

SUMMARY GRAPH

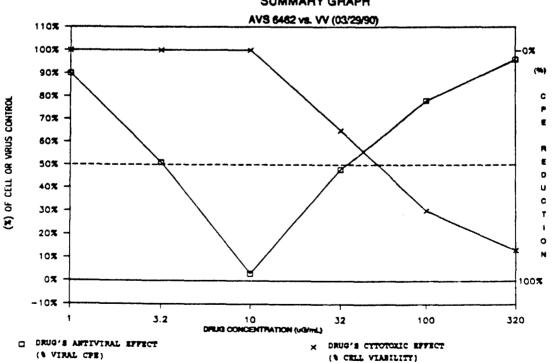


PLATE 0U2 DRUG 6462

# IN VITRO ANTIVIRAL RESULTS MTT ASSAY

DRUG: AVS 6462 TAI: 30.00 SI: 9.69

	1	2	3	4	5	6	7	8	9	10	11	12
ſ			reagent back	rground		1			pleastle backg	round		
A	0.125	0.117	0.119	0.120	0.124	0.130	0.000	0.000	0.000	0.000	0.000	0.000
Γ	494	0C/Y6	4ng	6462 expens	Mental	LOX .					00/49	
8	1.284	1.387	0.125	0.155	0.151	1.443	j			j	1.322	
c	1.197	1.494	0.453	0.332	0.473	1.315					1.435	
0	1.183	1.466	1.131	1.258	1.282	1.466					1.513	
εί	1.311	0.176	1.234	1.216	1.347	1.456	[			ſ	0.278	
F	0.415	6.175	0.359	0.399	0.430	0.446					0.187	
G	0.280	0.197	0.263	0.256	0.255	0.294					0.173	
Γ			drug 8462 00	continuents bec	aground							
н	0.124	0.106	0.110	0.108	0.103	0.152						
_	terment to					8010	- Disposi de					1

VIRUS	W
CETTS	VERO
SHIPMENT NUMBER	63
STRN	LEDCA
REAGENT	0.123
VIRUS CONTROL	0.075
CELL CONTROL	1.314
OIFFERENTIAL	1.238

	PROJECT #	5975-4
Satisfactory	SPONSOR	USAHRIID
CONFIRMS ORIGINAL ACTIVITY	TEST DATE	04/19/90
	DATE READ	04/25/90
	1 500	OFA

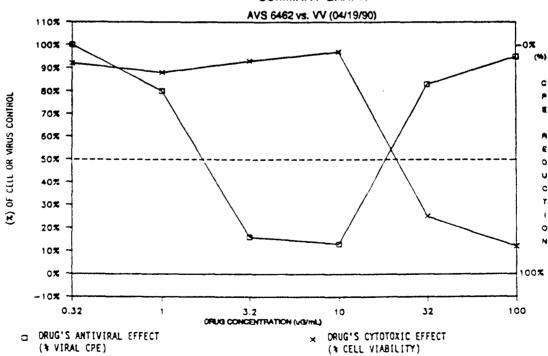
DRUG 6462	25%	504	954
TC (uG/mL)	16.70	24.40	> 100.00
IC (uG/mL)	1.10	1.72	
ANTIVIRAL INDEX (AI)	15.27	14.12	

DRUG	5452	ANTIVIRAL T	EST VALUES	CYTOTOXICI	TY TEST VALUES	·····
ROW ON PLATE	CONC. (uG/mL)	HEAN 0.D.	¥ VIRAL CPE	HEAN 0.0.	¥ CELL VIABILITY	COLORIHETRIC CONTROL
Tow B	0.32	084 0.242	1004 804	1.211 1.154	92 <b>%</b> 884	0.029
D E	3.2	1.041	164 134	1.217 1.274	934 974	015 013
F high G ≈	32 100	0.215 0.059	83 <b>4</b> 95 <b>7</b>	0.325 0.164	254 124	017 0.001

\* highest drug concentration tested

values shown are final adjusted numbers

### SUMMARY GRAPH



SOUTHERN RESEARCH INSTITUTE

	USAM	RIID	**************************************	
	Antiviral Drug Sc	reening Progra    SUBMITTER	CTR NO	05/18/90 AVS NO
TRUCTURE	CHIRAL	01141.01	KN-II-55	AVS-006466
		DATE RECD	AMT RECEIVED [mg]	MOL WT (au)
	0	12-28-89		224.218
		HANDLING/STORAGE		·
	NH			
•		SOLUBILITY		
0.	N O			
	$\checkmark$	STABILITY		
но				
	<b>=</b>	110 1111		
		ALT NAME	, 3'-DIDEOXYTHYMIDINE	NE
MPOUND NAME	2',3'-DIDEOXY	THYMIDINENE		
SCREEN IN	CTRICTION	TN V	IVO TOXICITY [n	ng/kg]
				(4) v 4 )
IORITY=PT>VEE>YF>KHF>PIC>JE	SF3VV3ADZ3VSV	HOST VM RTE SI	SO MTC LAS PR DATE	
	·			
IN VITRO SCRI	EEN [ug/ml]	IN VIVO	SCREEN [Dose	mg/kg]
VN VN+ 1050 CELL HTC V 4.99 HT2 40.0		VIR HST VR VR+ T	E KOT G STR HSV STR SEC	P L PR DATE
/C .32 CEM 51.5 NOT ACT VERO 64.3 NOT ACT VERO 111				
NOT ACT VERO 93.8 NOT ACT VERO 232	0 SO HTT 90-03-0	66		
NOT ACT VERO 100	0 SO MTT 90-03-0	)4		
	,			

PLATE 1HK

# IN VITRO ANTIVIRAL RESULTS MTT ASSAY

DRUG: AVS 6466 TAI: 35.67 SI: 9.78

	1	2	3	4	5	6	7		9	10	11	12
ſ			reagent back	rground					plante backg	round		
A	0.128	0.131	0.131	0.130	0.130	0.131	0.037	0.034	0.035	0.035	0.034	0.034
ſ	1046	99/19	drug	6488 expens	nen tel	lox				-	ac/va	
3	1.400	1.627	0.356	0.363	0.376	1.639					1.651	
c	1.362	1.625	0.418	0.386	0.384	1.622					1.588	
О	1.367	1.657	0.627	0.570	0.580	1.712				-	1.666	
2	1.430	0.369	1.821	1.729	1.850	1.785					0.358	
7	1.461	0.364	1.581	1.414	1.737	1.753					0.343	
9	0.160	0.368	0.127	0.130	0.142	0.156					0.338	
Ī			drug <b>64.66</b> 00	ionmetric bac	xground							
	0.133	0.129	0.131	0.135	0.131	0.132						
-						0010	- 51-5-04-40					

VIRUS	HIV3B
CELLS	MT2
SHIPHODE HUNGER	63
STRU	2.5
REAGENT	0.130
VIRUS CONTROL	0.227
CELL CONTROL	1.506
DIFFERENTIAL	1.279

Satisfactory; Active; Ret	•• <u>t</u>	SPONSOR TEST DATE	USAMRIID 04/04/90
		DATE READ	04/12/90
DRUG 6466	25%	50%	95%
TC (uG/mL)	48.80	66.40	97.90
IC (uG/mL)	3.53	4.99	9.33
APPIVIRAL INDEX (AI)	13.84	13.29	10.49

PROJECT #

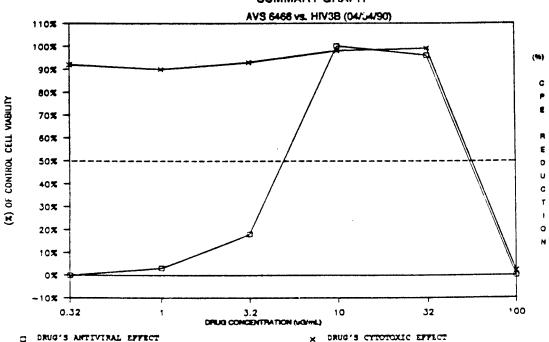
D.	RUG	6466	ANTIVIRAL	TEST VALUES	CALOLOXICI	TY TEST VALUES	
NOW	OH	CONC.	MEAN	* RED. IN	MEAN	• CZLL	COLORIMETRIC
PL	ATE	(uG/mL)	0.0.	VIRAL CPE	0.0.	VIABILITY	CONTROL
.04	3	0.32	0.006	0 🛰	1.387	924	0.002
	c	1	0.038	39	1.361	90%	0.001
	ם	3.2	0.231	189	1.404	939	0.005
		10	1.442	100	1.476	98%	0.001
	7	32	1.222	961	1.460	99%	001
ilgh	a -	100	227	0	0.025	2	0.003

\* highest drug concentration tested

values shown are final adjusted numbers

6520-2

#### **SUMMARY GRAPH**



DRUG'S ANTIVIRAL EFFECT
(% RED. IN VIRAL CPE)

DRUG'S CYTOTOXIC EFFECT

PLATE 1KA DRUG 6466

### IN VITRO ANTIVIRAL RESULTS MTT ASSAY

**DRUG: AVS 6466** TAI: >85.47 SI: >161.06

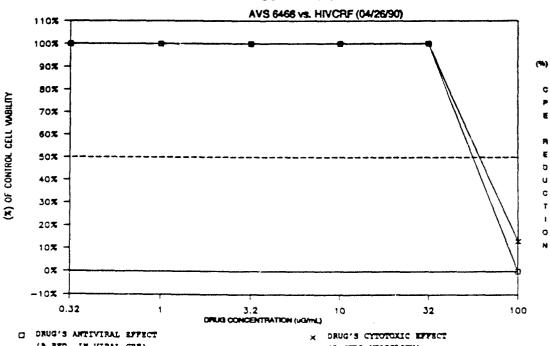
	1	2	3	4	5	6	7	•	9	10	11	12
Γ			segent beck	Brownd	**********				pleases becarg	raund		
A L	0.118	0.109	0.116	0.115	0.112	0.114	0.034	0.034	0.034	0.034	0.034	0.033
П		90/10					10x	drug	6466 expert	nontal	oc/ve	lox
3	Į.	1.009			1	į.	1.190	1.183	1.176	1.067	1.233	1.133
c		1.051			1	- 1	1.137	1.101	1.193	1.236	1.212	1.245
D	[	0.917			- 1	Ī	1.061	1.207	1.204	1.234	1.205	1.323
	ſ	0.478			ŀ	į	1.197	1.195	1.334	1.246	0.512	1.477
7	- 1	0.469			- 1	1	1.629	1.371	1.380	1.360	0.576	1.578
0	1	0.506			Ì		0.302	0.174	0.174	0.164	0.566	0.240
Γ							drug 8486 aplanmetric beckground					
						ĺ	0.144	0.137	0.135	0.126	0.121	0.132

VIRUS CELLS SEIPHONT NUMBER	HIVCRIF CEM 64	Satisfactory; Active; Ru Low MOI	<u>etest</u>	PROJECT # SPONSOR TEST DATE	6520-2 USAHRIID 04/26/90		
STR# REAGENT	RF2	Dama 4.44		DATE READ	05/03/90		
KENGERT	0.114	DRUG 6466	238	50%	95%		
VIRUS CONTROL	0.404	TC (uG/mL)	51.50	71.10	> 100.00		
CELL CONTROL	1.004	IC (ug/mL)	< 0.32	< 0.32	4 0.32		
DIFFERENTIAL	0.600	MPTIVIRAL INDEX (AI)	> 161.06	> 222.13	> 312.50		

ב	DRUG 6466		ANTIVIRAL	TEST VALUES	CYTOTOXICIT	TEST VALUES	
RO	ON	COMC.	HEAN	N RED. IN	MEAN	* CELL	COLORIMETRIC
PI	ATE	(uG/mL)	0.0.	CPE	0.0.	VIABILITY	CONTROL
low	3	0.32	0.606	1004	1.030	100%	0.018
	C	] 1)	0.652	100	1.070	1001	0.007
	D	3.2	0.685	100	1.066	100%	0.012
	I	10	0.720	100	1.202	100	0.021
	7	32	0.830	100	1.467	100	0.023
high		100	377	0	0.131	131	0.030

highest drug concentration tested

#### SUMMARY GRAPH



(\* RED. IN VIRAL CPE)

(% CELL VIABILITY)

PRINTED OUTSTHIPS

SOUTHERN RESEARCH INSTITUTE

					USAMI	RIID		
			P	ntivir	al Drug Scr	eening Progr	cam	05/18/
TRUC	TURE				CHIRAL	SUBMITTER 01141.01	CTR NO KN-II-95	AVS NO AVS-006467
						DATE RECD	AMT RECEIVED [mg]	MOL ST (au)
			HC1	NH		12-28-89	72.60	261.667
	•			//		HANDLING/STORA	GF.	
			N			MANUS 210.04	<b>95</b>	
			1					
	•	) HO	0-	` N		SOLUBILITY		<del></del>
	•		~ <i>₹</i> ~	>				
		'	\ \/			STABILITY		
		3	,,			SIMBILITI		
		НО						
						ALT NAME	ANHYDROCYTIDINE HYDROC	201.00125
						2,02-	ANTIDACCITIDINE HIDROC	CHLORIDE
MPOUN	D NAME			21 02		NE HYDROCHLORIDE		
					-ANNIDAGC11151.			
		SCREE	N INST	RUCTION		IN	VIVO TOXICITY [	mg/kg]
IORIT	Y-PT>VEE>Y	>KHF>P	IC>JE>SF	>VV>AD2>V	sv	HOST VH RTE	LD50 MTC LAB PR DATE	
	IN	VITRO	SCREEN	I [ug/m	11]	IN VI	VO SCREEN [Dose	= mg/kg]
	VR+ ID50	CELL	мтс		· LAB PRT DATE	VIR HST VR VR-	DOSE MTC VEH RTE D TOX	SP L PR DATE
v v	NOT ACT NOT ACT	HT2 HT2 VERO	.04 < .32 .71	0	SO HTT SO HTT SO HTT 90-03-2	2		
	NOT ACT	VERO VERO	1.8	0	SO MTT 90-03-0 SO MTT 90-03-2	<b>6</b> 2		
	NOT ACT NOT ACT NOT ACT	VERO VERO VERO	4.99 .50 2.4	0	SO MTT 90-03-0 SO MTT 90-03-2 SO MTT 90-03-0	2		
<u>:</u>	NOT ACT	VERO VERO	. 5	0	SO HTT 90-03-0 SO HTT 90-03-2	•		
	18 NOT ACT NOT ACT	VERO VERO VERO	. 82 . 52	10.53 0 0	SO MTT 90-03-2 SO MTT 90-03-2			
	NOT ACT	VERO	1.96	·	30 HIL 90-03-0	•		
						1		
							,	

	1	2	3	4	5	6	7	8	9	10	11	12
$\overline{}$			reagent back	ground					pleatic backg	round		
0	.105	0.097	0.123	0.114	0.109	0.119	0.000	0.000	0.000	0.000	0.000	0.000
		00/100					LOSE	grug	6467 experim		00/10	lox
l	- 1	1.314			1		1.488	0.153	0.166	0.134	1.321	1.330
1	- 1	1.339			1		1.510	0.179	0.148	0.205	1.470	1.42
		1.312			j		1.553	0.258	0.206	0.325	1.410	1.427
		0.182					1.415	1.153	1.398	1.302	0.160	1.54
1		0.180			ŀ		1.058	0.847	0.883	0.733	0.173	0.82
	[	0.174			ſ		0.450	0.426	0.446	0.427	0.142	0.400
									drug 6467 ool	orimetrio bao	kground	
1							0.107	0.110	0.105	0.108	0.114	0.117

VIRUS	w
CELLS	VERO
SHIPHENT NUMBER	63
STRM	LEDCA
REAGENT	0.111
VIRUS CONTROL	0.057
CELL CONTROL	1.250
DIFFERENTIAL	1.193

MONO CARD DEC	1 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2	OPA	
	DATE READ	04/25/90	
RETEST AT 3.2 UG/ML	TEST DATE	USAMR11D 04/19/90	
Satisfactory: Active: Retest	SPONSOR		
	PROJECT #	5975-4	

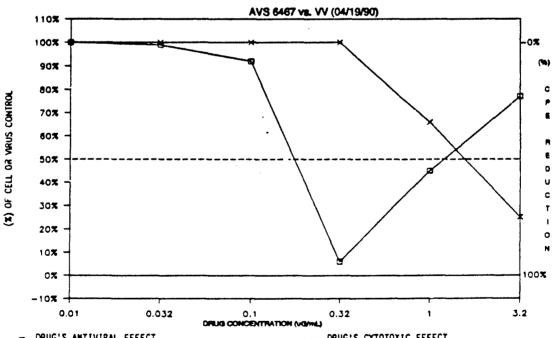
		DATE READ	04/25/90
DRUG 6467	254	504	954
TC (UG/ML) IC (UG/ML) ANTIVIRAL INDEX (AI)	0.82 0.13 5.52	1.86 0.18 10.53	3.20

	RUG	6467	ANTIVIRAL	TEST VALUES	CYTOTOXICI	TY TEST VALUES	
KON	ON	CONC.	HEAN	* VIRAL	HEAN	% CELL	COLORIMETRIC
PL	ATE	(uG/mL)	0.D.	CPE	0 <b>.</b> D.	VIABILITY	CONTROL
low	8	0.01	023	100%	1.252	100%	0.006
1	C	0.032	0.006	99%	1.353	100%	0.003
1	0	0.1	0.098	924	1.382	100%	003
1	E	0.32	1.122	64	1.373	100%	006
	F	1 1	0.654	45%	0.831	664	001
high	6 -	3.2	0.269	77%	0.318	25%	004

\* highest drug tencentration tester

values shown are final adjusted numbers





DRUG'S ANTIVIRAL EFFECT
(\* VIRAL CPE)

DRUG'S CYTOTOXIC EFFECT (\* CELL VIABILITY)

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